BULLETIN

OF THE

TORREY BOTANICAL CLUB

VOLUME 56

FOUNDED BY WILLIAM HENRY LEGGETT 1870

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NEW YORK
1929–30

PUBLISHED FOR THE CLUB
GEORGE BANTA PUBLISHING CO.
MENASHA, WISCONSIN

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Dates of publication

No. 1, for January, pages 1–64, issued 22 February, 1929
No. 2, for February, pages 65–122, issued 26 March, 1929
No. 3, for March, pages 123–180, issued 8 May, 1929
No. 4, for April, pages 181–226, issued 13 June, 1929
No. 5, for May, pages 227–284, issued 15 July, 1929
No. 6, for June, pages 285–338, issued 2 August, 1929
No. 7, for October, pages 339–390, issued 7 November, 1929
No. 8, for November, pages 391–448, issued 12 December, 1929
No. 9, for December, pages 449–587, issued 18 February, 1930

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VOL. 56 JANUARY, 1929 NO. 1

BULLETIN
OF THE
TORREY BOTANICAL CLUB

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PUBLISHED FOR THE CLUB MONTHLY
EXCEPT JULY, AUGUST AND SEPTEMBER

BY

GEORGE BANTA PUBLISHING COMPANY
MENASHA, WISCONSIN

SUBSCRIPTION PRICE PER YEAR $4.00

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Studies on the flora of northern South America—XI.[1]

New or noteworthy monocotyledons from British Guiana

H. A. Gleason

The genus *Carludovica* contains at least thirty, and probably a good many more, species of tropical America. They are either acaulescent and palm-like in aspect, or climbers with palm-like leaves, and have a very characteristic and unusual type of flower-structure. The genus dates back to Ruiz and Pavon, who gave in 1798 only the briefest mention of the structural features of the flower, and not enough to enable one to relate their species to modern groups, except in the case of the economic species *C. palmata*. In 1807 Persoon, objecting to the length of the name, changed it to *Ludovia* and transferred the original five species. Oersted in 1857 proposed the genera *Sarcinanthus* and *Evodianthus*, described them well, and later gave excellent illustrations. Only one species has been associated with the former name and but three with the latter. They were merged with *Carludovica* by Bentham and Hooker, but kept separate by Drude in the Pflanzenfamilien. Brongniart in 1861 based a new genus on a Guiana plant and gave it the name *Ludovia*, since Persoon's *Ludovia* had never come into use. Drude founded a fifth genus *Stelostylis*, with one species, in 1881.

In the meantime numerous species had been described under the broad generic concept of *Carludovica*, and in probably half of them the structure of the flowers was not mentioned, or was stated in the most general terms; many of the species were based on completely sterile material, or on living plants in ornamental collections, the identity of which is notoriously easily lost. As a result, of about sixty species of *Carludovica* which have been proposed, at least half are little better than *nomina nuda*.

[1] Contributions from the New York Botanical Garden, No. 305.

[The Bulletin for December (55: 479–570) was issued 24 January 1929.]

The omission of observation of flower-structure by so many botanists is difficult to account for. Poiteau, as early as 1822, gave a description and illustration of one species, the structure of which is so remarkable that it should have excited every contemporary to verify it and to confirm it among other species. Hooker gave excellent plates of species built on a plan so difficult that it is strange that they escaped segregation as genera. Yet Drude in the Flora Brasiliensis mentions the structure of the staminate flower in only three species out of fourteen, and a few years later described the genus in the Pflanzenfamilien in the most general terms only.

Material from British Guiana shows six general types of flower-structure, each sufficiently different to be ranked as a genus, as indeed two of them have. It seems better, however, to keep them in all in *Carludovica* as sections, where they are just as valuable, or more so in our present lack of knowledge of so many other species. If several new genera are to be segregated, there will be little left of *Carludovica* except the Panama-hat plant and its allies, and the remaining species can seldom be assigned with any degree of accuracy to a new genus because of lack of proper material for study.

These six subgeneric groups include eight species in British Guiana; others occur in Trinidad, Surinam, and French Guiana, representing in some cases still other subgenera. The association of these eight species with any of the sixty already described is very difficult and has been accomplished in only two cases. The subgeneric characters are presented in the key:

Receptacle of the staminate flowers flat or concave, the pedicel attached excentrically, distinctly asymmetric, the adaxial side more or less truncate, the remainder of the margin with 5–8 spreading lobes; stamens 5–12, on the upper surface, mostly marginal; spathes 2 or 3, all at the summit of the peduncle, the outer one or two lanceolate to ovate, the innermost enclosing the spike before anthesis and abruptly contracted above into a linear cusp; small or delicate acaulescent plants with spadices 1–2 cm. long . . . . . Section I.

Receptacle of the staminate flowers funnelform or deeply concave, bearing the numerous (25–50)stamens on the inner surface; perianth-lobes 10 or more, symmetrically disposed around the margin.

Perianth-segments erect, somewhat enlarged distally, terete or radially flattened, hollow, imbricate in two series of which the outer is the smaller, with short, acute, distinctly inflexed tips; spathes 3 or 4, all above the middle of the peduncle ..... Section II.

Perianth-segments tangentially flattened, rather thick and fleshy, regularly imbricate in one series.

Sepals of the pistilllate flower short-triangular, equaling the sessile stigmas, all about equal in size; perianth-segments of the staminate flower about 12, more than 1 mm. wide, decurrent on the outside of the receptacle; stamens about 50–60, linear-oblong, 1.2–3 mm. long; spathes scattered, 8–10 ..... Section III.

Sepals of the pistillate flower rhomboid, one opposite pair twice as wide as the other, erect; style elongate, stoutly pyramidal, 3 mm. long, the cruciform stigmas not wider; perianth-segments of the staminate flowers minute, not decurrent; stamens about 25, short-oblong or almost square, about 0.5 mm. long; spathes 3, near the middle of the peduncle, the spadix stipitate ..... Section IV.

Receptacle of the staminate flowers flat or nearly so, the pedicel expanded above into a solid obconic flat-topped receptacle, bearing the numerous stamens scattered over its surface; perianth-lobes marginal, small, in one or two series or none.

Receptacle of the staminate flowers nearly or quite symmetrical; filaments waxy-white, strongly swollen and subglobose at base ..... Section V.

Receptacle asymmetric, the abaxial side with 4–7 large semiglobose submarginal yellow glands; perianth-lobes none; filaments slender ..... Section VI.

SECTION I. Herbaria contain other specimens which greatly resemble the three following in general aspect and which doubtless belong also to this section.

Receptacle of the staminate flowers flat or nearly so, its lobes 4–10, those on the abaxial side linear or narrowly triangular; filaments very short and stout, or the anthers essentially sessile, with widely divergent pollen-sacs.

Filaments 0.2 mm. long; perianth-lobes finger-shaped, obtuse; stamens 4–9 ..... *C. pygmaea.*

Filaments none; perianth-lobes narrowly triangular, acute; stamens 12 ..... *C. insularis.*

Receptacle of the staminate flowers somewhat concave, or becoming flattened with age, its lobes about 8, short, obtuse; stamens 12; filaments short but very slender; pollen-sacs parallel and approximate..... *C. nana.*

Carludovica pygmaea n. sp. Terrestrial, the leaves crowded on a short caudex about 5 cm. high; petioles slender, 6–10 cm. long, somewhat expanded below, mingled with linear sheaths 4–5 cm. long; leaf-blades 11–18 cm. long, bipartite for four fifths of their length, with a single common midrib, the segments linear-lanceolate, 10–13 mm. wide, long-acuminate; peduncles slender, several on each caudex, terete, about 7 mm. long; spathes 2, apical, the lower triangular, 20–25 mm. long, 7–10 mm. wide near the base, gradually tapering into a long slender tip, the upper enclosing the spadix before anthesis, its body subrotund, abruptly contracted distally into a linear cusp 5–7 mm. long; spadix 10–15 mm. long; pistillate flowers sessile, the 4 perianth-segments depressed-ovate, nearly concealed, bearing filiform staminodes; stigma cruciform, sessile, each lobe obovate; staminate flowers on short flattened pedicels; receptacle abruptly expanded, flat, somewhat circular, inequilateral, 1.8–2.3 mm. wide, truncate along the smaller adaxial side, elsewhere bearing 5–9 obtuse, finger-shaped lobes 0.4–0.6 mm. long; stamens 1 central and 3–8 near the margin; filaments very stout, 0.2 mm. long, prolonged into the stout connective so that the oblong pollen-sacs (0.6 mm. long) become widely divergent at base.

Type, *Hitchcock 17,388*, collected in forest at Tumatumari, British Guiana, 3–5 Jan. 1920, and deposited in the herbarium of the New York Botanical Garden.

Carludovica insularis n. sp. Plants larger and stouter than the preceding; terrestrial; petioles as much as 35 cm. long; leaf-blades 50 cm. long, bipartite for three fifths of their length, with a single common midrib, the segments 5 cm. wide, not notably acuminate; peduncles slender, as much as 3 dm. long, mingled below with linear sheaths; spathes 2, apical, the outer 8 cm. long, ovate-lanceolate below, narrowed above into a linear cusp 5 cm. long, the inner broadly ovate-rotund, closely enwrapping the spike before anthesis, abruptly narrowed into a short linear cusp 8 mm. long; spadix 15–20 mm. long; pedicel of the staminate flowers flattened, abruptly dilated into a flat receptacle, the latter somewhat pentagonal in shape, about 3.5 mm. in diameter, truncate on the narrower side, elsewhere irregularly 7–9-lobed, the lobes triangular-linear, acute, 1–1.5 mm. long; stamens 12, some central, the majority near the margin or even on the ends of the lobes; anthers sessile by a

broad base, the pollen-sacs oblong, 0.5–0.6 mm. long, approximate above, widely divergent below.

Type *Broadway 4066*, collected in woods, Tobago, British West Indies, 30 Sep. 1911, and deposited in the herbarium of the Royal Botanic Gardens in Kew. A second sheet, purporting to be the same collection, has a huge leaf with 3 midribs, irregularly lobed above. It is very doubtful if it actually belongs here. Another Tobago sheet of the species, collected two years later, is much smaller in every way, the petioles and peduncles about 1 dm. long and the blades 17–21 cm. long.

Carludovica nana n. sp. Acaulescent, terrestrial; petioles dilated at base, slender, 17–25 cm. long, more or less mingled with linear sheaths; leaf-blades 15–20 cm. long, bipartite for four fifths of their length, with a single common midrib, the segments 15–20 mm. wide, slenderly acuminate; peduncles slender, about 2 dm. long; spathes 3, apical, the two outer lanceolate, 5 and 4 cm. long, slenderly acuminate, the inner broadly ovate at base, closely enfolding the spike, abruptly narrowed to a cusp 8 mm. long; spadix 15 mm. long; staminate receptacle broadly and shallowly funnel-form, becoming somewhat explanate in age, about 1.5 mm. in diameter, asymmetric, the narrower side truncate, the remainder of the margin bearing about 8 short, obtuse, irregular lobes; stamens 12, mostly near the margin, the pollen-sacs broadly oblong, 0.4–0.6 mm. long, parallel and opposite on very slender filaments 0.4 mm. long.

Type collected by *R. H. Schomburgk*, without number, in stony beds of rivers and brooks in the Serra Mey, British Guiana, and deposited in the herbarium of the Royal Botanic Gardens at Kew.

SECTION II. Only one species has been described in such terms that it can be authentically referred to this section, *C. funifera* (Poit.) Kunth, a plant of French Guiana, with leaves which are bifurcate only in the distal third, according to Poiteau. To this we now add a second:

Carludovica coronata n. sp. Stems stout, climbing to a height of 15 m.; petioles 15–30 cm. long, sheathing at base, channeled on the upper side; leaf-blades 3.5–6.5 dm. long, bifurcate for two thirds to four fifths of their length, or rarely less, with a single common midrib, the segments many-veined, acuminate, 25–50 mm. wide; peduncles 15–20 cm. long; spathes 2–4, at or near the summit, ovate to ovate-lanceolate, acumi-

nate, soon deciduous, exceeding the spike; spadix 3–5 cm. long; staminate flowers obconic on a short pedicel, about 4 mm. in diameter, perianth-lobes about 20, biseriate, terete or slightly flattened radially, hollow, rounded on the top, the actual tip acute and inflexed, the inner series somewhat obovoid, the outer similar but smaller; stamens about 24, in two circles of about 12 each, on the inside of the deeply concave receptacle; anthers oblong, 0.6 mm. long, about equaling the slender filaments.

Type, *De La Cruz 4444*, collected at Kaieteur Falls, British Guiana, 23 Oct.–3 Nov. 1923, and deposited in the herbarium of the New York Botanical Garden. The species is widely distributed through British Guiana, and is represented at Kew or New York by *Appun 259*, *Jenman 736*, *1317*, *Hitchcock 16,988*, *Gleason 150*, *De La Cruz 1445*, *1934*, *3285*, and the type.

SECTION III. The manuscript name of *C. sarmentosa* was was applied by Sagot to a specimen from French Guiana, but was never published by him. Drude published it for Sagot, citing only Brazilian material and possibly drawing his description from that rather than from the Guiana plant. A specimen at Kew of the original collection enables us to refer the species to this group.

Spathes broadly rotund to subcordate at base..... *C. sarmentosa.*
Spathes narrowed to the base..... *C. bracteosa.*

Carludovica bracteosa n. sp. Stems climbing; petioles about 1 dm. long, flattened or channeled above; leaf-blades tripartite for about two thirds of their length, inequilateral, the single midrib running to one sinus, the single segment 2–3 cm. shorter at base than the double segment, all segments 3–5 cm. wide, acuminate, 7–10 costate, somewhat plicate; peduncles stout, 15 cm. long; spathes about 10, evenly distributed, the lowest lanceolate, 2 cm. long, the upper enlarged, linear-oblong, 8–10 cm. long, acute, narrowed to the base; spike ovoid, 4–5 cm. long; perianth-segments of the pistillate flowers erect, triangular, acute, 3 mm. long and wide, equaling the cruciform stigmas; staminodes about 3 cm. long; staminate flowers campanulate, on a pedicel 3.5 mm. long, the hollow interior 2.7 mm. deep; perianth segments 12, 3 mm. long, only the distal 1 mm. free, the whole segment linear-oblong, decurrent on the receptacle, contiguous below, abruptly dilated at the free tip to 1.5–1.7 mm. wide and half overlapping with adjacent segments, rounded or barely retuse at the fleshy pinkish tip;

stamens about 60, covering the inside of the flower; filaments slender, 0.8 mm. long; anthers erect, linear-oblong, the central 2.7 mm., the others progressively reduced to 1.2 mm. long.

Type, *De La Cruz 1505*, collected at Dadanawa, British Guiana, 14 June 1922, and deposited in the herbarium of the New York Botanical Garden; a second sheet is *De La Cruz 1309*.

C. Kegeliana Lem., described as having ten spathes, may also belong to this section.

SECTION IV. **Stelostylis** (Drude, as genus). The essential feature of Drude's genus is the elongate style, but it may be distinguished, at least from Guiana species, equally well by staminate structures. The following differs from Drude's *Stelostylis coriacea* in its long petioles and peduncles, prominently veined leaves, and 25 stamens.

Carludovica stylaris n. sp. Petioles 6 dm. long; leaf-blades 7 dm. long, bifurcate for three fourths of their length, with a single common midrib, the segments 6 cm. wide, abruptly acuminate, prominently 7–8-nerved; peduncles 2–2.5 dm. long, stout, slightly angular, invested at base by 2–3 elliptical obtuse or acute sheaths 4–5 cm. long; spathes apparently 3, the lower somewhat beyond the middle, the upper 2–3 cm. below the spike, narrowly lanceolate, 8 cm. long; spadix becoming 5 cm. long at maturity; perianth-segments of the pistillate flowers thick and woody, 3 mm. long, 1.5 or 3 mm. wide, obtuse; style stout, pyramidal, 4-angled, 3 mm. long, the cruciform stigmas 2 mm. in expanse; staminate flowers funnelform or obconic, 2 mm. long, fleshy, the receptacle concave within; perianth-segments about 12, erect with recurved margins, semicircular, fleshy and rounded, 0.3 mm. long, 0.4–0.6 mm. wide; stamens about 25, covering the receptacle; anthers nearly square, 0.5 mm. long, flattened tangentially, on slender filaments 0.2 mm. long.

Type, *Jenman 898*, collected on trunks of trees and rocks, in forests near the Kaieteur savanna, British Guiana, Sep.-Oct., 1881, and deposited in the herbarium of the Royal Botanic Gardens at Kew. Field notes state that the stem is two inches thick and not climbing.

SECTION V. **Sarcinanthus** (Oersted, as genus). A sheet of *Jenman 900*, from British Guiana, matches nicely with the type collection of *Carludovica latifrons* Drude and is referred to

that species, while its flower structure shows clearly that it is related to Oersted's species and to the few others that have elsewhere been described with similar characters.

SECTION VI. I know of no other published species with which the following may be compared.

Carludovica glandulosa n. sp. Terrestrial, acaulescent; petioles slender, 4 dm. long, channeled and wing-margined throughout, very gradually expanded into the blade; leaf-blade about 11 dm. long, bifurcate, the segments 14 cm. wide, acuminate, many-veined; peduncles 8–10 cm. long, bearing above the middle 3 ovate-oblong, sessile and clasping, acuminate spathes 5–6 cm. long; spadix on a stalk 1 cm. long, ellipsoid or cylindric, 25–30 mm. long; staminate flowers woody, asymmetric, flat on top, 2.2 mm. in diameter, obconic; perianth-segments none; receptacle bearing on the outer side, just below the margin, 4–7 semiglobose yellow glands, the largest 0.6 mm. in diameter, the smaller ones at the ends of the row half as large; stamens about 25; anthers oblong, 0.6 mm. long by 0.4 mm. wide, on slender filaments 0.2 mm. long.

The type is *Jenman 2105*, collected in the Mt. Russell District, British Guiana, and deposited in the herbarium of the Royal Botanic Gardens at Kew.

Anthurium expansum n. sp. Section *Schizoplacium:* epiphytic, probably scandent, the stem stout and thick; leaf-blades ample, thinly membranous, 7-parted; central three lobes separate to the base, elliptic-oblong, 35–40 cm. long by 12 cm. wide, gradually narrowed above to a slender cusp 2 cm. long, long-cuneate below to a sessile base; two lateral lobes on each side connate for about 8 cm. above the inequilaterally cuneate base, the inner lobe narrowly elliptic-oblong, 35 cm. long, 8 cm. wide, the outer lobe falcate-lanceolate, 28 cm. long by 4.5 cm. wide, inequilateral at base and slenderly acuminate; costae elevated on both sides, the lateral veins 8–12 mm. apart, plane or somewhat depressed above, prominulous beneath, ascending at an angle of 50–60° and confluent into a collective nerve 2–3 mm. from the margin; peduncle at least 3 dm. long; spadix narrowly conical, 15 cm. long; spathe linear-lanceolate, equaling the spadix.

Type, *Lang 223*, collected at Kurupung, Mazaruni River, British Guiana, 23 Nov. 1922, and deposited in the herbarium of the New York Botanical Garden. *Anthurium expansum* appears to be most closely related to *A. Andersonii* Schott,

and differs in its much larger, 7-parted leaves with elliptic lobes, in its submarginal collective nerve, and in its larger spathe and spadix.

Anthurium glaucophyllum n. sp. The name is proposed to cover a plant from British Guiana, cultivated at Kew, where it flowered in October, 1891, and where type material is conserved in the herbarium. Petioles slender, 4 dm. long; leaf-blades oblong-lanceolate, thin but firm, 5 dm. long, 13 cm. wide, acute, broadly rounded at base, very glaucous beneath; lateral veins numerous, 1–2 cm. apart, ascending at an angle of 60°, veinlets prominent beneath, conspicuously reticulate above; collective nerve prominent, about 1 cm. from the margin; peduncle slender, 5 dm. long; spathe narrowly ovate-oblong, 4 cm. long, erect, equaling the slender spadix.

Anthurium potarense n. sp. Section *Semaeophyllium;* stem not known; petiole slender, 3–4 mm. in diameter, subterete, channeled on the upper side, 4–5 dm. long (or more); blades firm in texture, drying brownish green, deeply 5-lobed, the central three lobes narrowly obovate-oblong, 7–11 cm. wide, 32–40 cm. long, gradually tapering to an abruptly acuminate tip, long-cuneate at base, the outermost lobes narrowly oblong, 27–30 cm. long, 5–6 cm. wide, acuminate, connate with the interior lobes for 1–2 cm., equilateral except for a semicircular basal lobe about 6 cm. in diameter; lateral veins prominent, arising at an angle of about 75°, slightly arcuate and confluent into a collective nerve 3–5 mm. from the margin; veinlets prominently reticulate; peduncle at least 2 dm. long; spathe narrowly lanceolate, about equaling the spadix, which is slenderly cylindric, 10 cm. long.

Type, *Jenman 7466*, collected on the Potaro River, British Guiana, Oct. 1898, and deposited in the herbarium of the Royal Botanic Gardens at Kew. *A. potarense* seems to be most closely related to *A. fissum* C. Koch, but the leaves are much more deeply lobed.

Anthurium obovatum n. sp. Stems elongate, climbing by rootlets, 2 cm. in diameter, the internodes 3–5 cm. long; petioles 6–7 dm. long, winged along the upper side for 15 cm. at the base, the remainder slender, channeled above; leaf-blades 7-parted, rather thin but firm; middle 3 segments distinct to the base, obovate, 4–4.5 dm. long, 16–17 cm. wide, abruptly rounded above to a falcate apiculum 1 cm. long, narrowed below the middle to a long-cuneate, petiole-like, winged base; lateral veins 10–20 mm. apart, ascending at an angle of 60° and very slightly arcuate, connected by a collective nerve about 5 mm.

from the margin, elevated on both surfaces; veinlets prominulous beneath and conspicuously reticulate; fourth and fifth segments resembling the preceding and only slightly smaller, their base more broadly winged and connate with the outermost segments for 4 cm. above the base, their midribs not confluent; outermost segments narrowly obovate, 35 cm. long, 12 cm. wide, apiculate, the inner margin long-cuneate below to the point of confluence with the inner segment, the outer margin with a thumb-like basal projection, its free limb oblong, apiculate, 10 cm. long above the narrow sinus, 6 cm. wide, traversed by the usual collective nerve and by a costa which leaves the midvein of the segment 6 cm. above its base and is denuded on the outer side for 8–9 cm. above the summit of the petiole, its outer margin broadly rounded below to a cuneate base; peduncle stout, 3 dm. long, oval in section; spathe linear-oblong, 2 dm. long, 3 cm. wide, apiculate, conspicuously veined; spadix stout, purple, 2 dm. long, 2 cm. in diameter as pressed.

Type, *Altson 475*, on tree trunks in the forest, Anandabaru, Kopinang River, British Guiana, April, 1926, deposited in the herbarium of the Royal Botanic Gardens at Kew. It is a member of the section *Schizoplacium* and differs from the related species in the size of the spathe and spadix and the characters of the leaf.

Dieffenbachia paludicola N. E. Brown. Perhaps stemless; petiole 20 cm. or more long, and in the dried specimen 2 cm. thick; blade cuneately oblanceolate, acute, uniformly green, about 75 cm. long and 18 cm. broad at about 25 cm. below the apex, thence gradually narrowing into the petiole; midrib stout; primary lateral veins about 12, curved, ascending; peduncle about 30 cm. long, and in parts of the dried specimen 8 mm. thick, so probably much stouter when alive; spathe about 32 cm. long and in the dried state about 2 cm. in diameter, shortly acute at the apex, apparently green; adnate part of the spadix about two-thirds as long as the spathe, with the fertile and barren flowers laxly scattered along it; ovary 2–3-celled; male part of the spadix not seen.

British Guiana: in a swamp near the Penal Settlement, *Hitchcock 17030* (type in herb. New York Botanical Garden). In all probability *Jenman 5771* is the same species, but the specimen at Kew is too imperfect to decide, only the upper part of the leaf being represented. It was collected on the Essequibo River and is stated to be 'acaulescent, gregarious in water in forests, no clear petiole.'

Philodendron demerarae n. sp. Stems acaulescent, epiphytic; peticles stout, not winged, 3–6 dm. long, 1 cm. wide or less when pressed; leaf-blades cordate-sagittate, the margin at the base distinctly ascending from the summit of the petiole to form a broadly cuneate base, thence at once spreading or somewhat recurved into the basal lobes, which are semi-circular or depressed ovate, broadly rounded, and somewhat divergent; terminal lobe triangular-ovate, somewhat longer than broad, acute to abruptly acuminate; lateral veins from the midvein 12–18 on each side, the upper straight and ascending at an angle of about 60°, the lower 3–5 approximate at base, spreading at right angles or recurved and gradually diverging in the basal lobes, the lowest veins not naked in the sinus; lateral veins of the second rank fine, closely parallel, about 1 mm. apart; peduncle apparently 5 cm. long, slender; spathe 7–10 cm. long, purple at base, white above, closely convolute below; spadix about 7 cm. long, of which the basal fourth is pistillate and scarcely swollen, and the terminal two-thirds is staminate; ovules numerous in a vertical row in each loculus.

Type, *Hitchcock 16,987*, collected on trees in wet forest southeast of Lamaha Stop-Off, British Guiana, Nov. 27, 1919, and deposited in the herbarium of the New York Botanical Garden. Other sheets referred here are *Jenman 5759* from the Essequibo River and *Gleason 121* from Tumatumari. The leaves measure 32, 42, and 44 cm. along the midvein of the terminal lobe by 23, 35, and 41 cm. wide.

Philodendron platypodum n. sp. Stem slender, climbing, the internodes 2–3 cm. long; petioles stout, 8–10 cm. long, broadly winged to the summit, 15 mm. wide at base, 10 mm. at the rounded apex; leaf-blades firm in texture, elliptic, distinctly inequilateral, 21–23 cm. long, 9.5–11 cm. wide, abruptly narrowed above to a cusp 1 cm. long, broadly rounded at base; principal lateral veins 6–9 on each side, ascending at an angle of 70°, scarcely more prominent than the secondary, which are 1 mm. apart; peduncle stout, 2.5 cm. long; spathe 12 cm. long, green within and without, closely convolute at base, not obviously constricted, partially expanded and cuspidate above; spadix 13 cm. long.

Type, *Jenman 5762*, collected at Lama, British Guiana, Apr. 1889, and conserved in the Jenman Herbarium. The species is a member of the section *Pteromischum*, and finds its nearest relative in *P. cannifolium* (Rudge) Engler, which differs in its narrower, thinner leaves, cuneate to the base and more distinctly acuminate, and in its cream-colored, spotted spathe.

Philodendron subsessile n. sp. Stems slender, climbing, the flexuous internodes 2.5–5 cm. long, the nodes marked by lanceolate cataphylls 2 cm. long; petioles slender, wingless, 5–15 mm. long; leaf-blades thin, dark green, cordate-ovate, 5–9 cm. long, 3–7 cm. wide, abruptly acuminate into a short filiform cusp 1–2 mm. long, the basal lobes rounded, separated by a narrow sinus about equaling the petiole; lateral veins about 6 on each side, poorly developed, more or less approximate toward the base of the leaf.

Type, *Gleason 460*, collected in dense forest at Tumatumari, British Guiana, 18 June–8 July 1921, and deposited in the herbarium of the New York Botanical Garden. Other specimens from Guiana are *Gleason 461*, *De La Cruz 1029*, and *De La Cruz 3708*. These all have smaller and proportionately narrower leaves, but differ from the type in no essential respect.

Philodendron dioscoreoides n. sp. Stems slender, climbing, the comparatively straight internodes 5–7 cm. long; cataphylls at the nodes apparently none; petioles 3–6 cm. long, nearly equaling the blade, conspicuously winged on the basal half or nearly to the summit; leaf-blades narrowly cordate-ovate, 6–9 cm. long, 4–6 cm. wide, acuminate into a slender cusp 2–3 mm. wide and about 1 cm. long, often falcate, the basal lobes rounded, separated by a broad shallow sinus; principal lateral veins poorly developed, about 6 on each side.

Type, *Gleason 459*, collected in dense forest at Tumatumari, British Guiana, 18 June–8 July 1921, and deposited in the herbarium of the New York Botanical Garden. Other specimens are *Bartlett 8125* and *Gleason 449*, both of which have smaller leaves than the type.

P. subsessile and *P. dioscoreoides* both represent sterile species, possibly juvenile, or possibly sterile because of some environmental deficiency. Both were common in the forest at Tumatumari, but repeated search failed to reveal any connection between either of them and other fertile species. Both are related to *P. micans*.

Rhodospatha venosa n. sp. Stem high-climbing, with both aerial and hold-fast roots, woody, the internodes about 12 cm. long by 1 cm. in diameter; petioles slender, vaginate to the geniculus, which is 15 mm. long; blades membranous, green on both sides, ovate-oblong, 33 cm. long by half as wide, abruptly acute, rounded at base; principal lateral veins 1 cm. apart, scarcely more prominent than the secondary (1–1.5 mm.

apart), ascending at an angle of 45–60°; leaves on the peduncle 3, bladeless, strongly vaginate, narrowly linear, 13 cm. long, borne 6 cm. below the summit; spathe not seen; spadix cylindric, obtuse, 8 cm. long, on a stipe 1 cm. long; flowers all perfect.

Type, *Gleason 153*, from dense upland forest, Tumatumari, British Guiana, in the herbarium of the New York Botanical Garden. It is apparently related to *R. Melinoni* (Engl.) Engl. and Krause, in which the leaf is glaucous beneath, with the veins leaving the midrib almost at right angles.

Syngonium connatum n. sp. Leaf-blades conspicuously bluish-green above, dull green beneath, deeply 3-parted; terminal segment elliptic to rhombic-elliptic, 25–40 cm. long, 13–15 cm. wide, abruptly short-acuminate, equilateral, narrowed to a broadly sessile base; lateral segments ovate, 20–30 cm. long, 9–14 cm. wide, acute, strongly inequilateral or somewhat falcate, the upper side two-thirds as wide as the lower, concavely cuneate at base into a wing 1 cm. wide confluent with the central segment, the lower side broadly rounded or subcordate to an abruptly cuneate base, the stalk naked for 15–20 mm.; all lateral veins nearly parallel, straight or slightly arcuate below, curved forward near the margin, and finally confluent, but without forming a definite collective nerve.

Type, *Jenman 5756*, collected along the Rupununi River, British Guiana, Oct. 1889, and deposited in the herbarium of the Royal Botanic Gardens at Kew. Only the leaf-blades are available on the three sheets examined. The great size, bluish or glaucous tint, and broadly winged base of the lateral leaflets distinguish it from all other species seen by me.

Syngonium ternatum n. sp. Stems climbing, juice milky; petioles about 3 dm. long, conspicuously wing-margined for about half their length; leaf-blades membranous, bright green, trifoliate; terminal segment nearly sessile, equilateral, oblong, 17–18 cm. long, 6–7 cm. wide, obtuse, narrowed to the base, the lateral veins ascending irregularly at an angle of about 45°, united into an obscure collective nerve about 8 mm. from the margin; lateral segments elliptic, inequilateral or somewhat falcate, as long as the terminal, obtuse or subacute, the inner half cuneate to an acute base, the outer half narrowed to an obtuse or slightly cordate base; stalks of the lateral segments 10–15 mm. long, naked on the basal side, very narrowly winged on the distal; peduncles axillary, about 1 dm. long, clavate above; spathes and spadix not seen; the syncarpous berry yellow, 3 cm. long.

Type, *De La Cruz 1288*, collected along the Waini River, British Guiana, 6 Feb. 1922, and deposited in the herbarium of the New York Botanical Garden. *De La Cruz 1469*, from the upper Rupununi River, is also referred here: it has leaves 22 cm. long, the lower half of the lateral segments subcordate at base, and a fruit with about 10 carpels. The species most nearly resembles *S. yurimaguense* Engl. from Peru, which has smaller and proportionately narrower leaflets with a rather prominent basal lobe, and petioles winged for three-fourths of their length.

Paepalanthus Jenmani n. sp. Caudex very short, thinly lanate; leaves densely cespitose, soft and lax, erect, narrowly linear, 1–2 mm. wide, 8–12 mm. long, glabrous, subulate-tipped; peduncles 20–25 cm. tall, roundly 3-costate, glabrous, twisted, their sheaths strongly twisted, sparsely hirsute, 4–5 cm. long, with long-oblique orifice and subscarious lamina; heads hemispheric, 3–5 mm. wide; involucral bracts broadly ovate to ovate-oblong, appressed, regularly imbricate in several series, glabrous; receptacle hairy; staminate flowers: pedicel very slender, glabrous, 1.3–1.7 mm. long; perianth hyaline; sepals free, glabrous, elliptic-obovate, obtuse, 1.7 mm. long; petals free, narrowly obovate, 2 mm. long, obtuse or rounded at apex, glabrous; stamens 3, inserted on the very base of the corolla, opposite the petals, about equaling the sepals, the filaments slender, the anthers 2-locular; pistillate flowers sessile; sepals free, elliptic, 1.5 mm. long, obtuse, glabrous; petals subulate-spatulate, erect and closely appressed to the pistil, 2.3 mm. long, 0.1–0.2 mm. wide, densely hirsute throughout with erect hairs; staminodes none; ovary glabrous, thick-fusiform 0.5–0.6 mm. long; style very short; stigmas 3, filiform, simple; appendages 3, inserted at base of stigmas, erect, narrowly clavate, just equaling the stigmas.

Type, *Jenman 1032*, collected on the Kaieteur savanna, British Guiana, Sep.–Oct. 1881, and deposited in the herbarium of the Royal Botanic Gardens at Kew. It is most closely related to *P. capillaceus* Klotzsch, from which it may be distinguished by its wider leaves and narrower, densely hirsute petals in the pistillate flowers.

Syngonanthus brevifolius n. sp. Stems tufted, freely branched, 3–5 cm. long, somewhat lanate, leafy in the terminal centimeter; leaves crowded, recurved or spreading, narrowly lanceolate, 4–6 mm. long, 1 mm. wide, glabrous, prominently nerved; peduncles solitary in some of the upper axils, 10–16

mm. long, slender, 3-angled, somewhat twisted, sparsely hirtellous and occasionally glandular; sheaths ample, 3–4 mm. long, glabrous, obliquely truncate at the summit to form an obtuse lamina 1.2 mm. long; heads 2–3 mm. in diameter; outer bracts about 5, broadly oblong-elliptic, 1.1–1.5 mm. long, 0.9–1.1 mm. wide, glabrous or nearly so, membranous, rounded at apex; inner bracts oblong, obtuse, 1.2 mm. long by 0.5 mm. wide, glabrous; pistillate flowers: pedicel 0.7 mm. long, hirsute; sepals scarious, obovate-oblong, symmetrical, 1.0 mm. long by 0.5 mm. wide, rounded at apex, glabrous; petals separate, scarious, spatulate-obovate, 1.2 mm. long, 0.5 mm. wide, glabrous, rounded at the apex; style 0.2 mm. long, bearing 3 simple stigmas without appendages; staminate flowers: sepals scarious, cuneate-obovate, 1.0 mm. long, 0.4 mm. wide, glabrous, rounded at apex; petals connate into a broadly obconic tube 0.7 mm. long, not involute, crowned with 3 lobes 0.15 mm. long; stamens exserted.

The type is *Jenman 4755*, collected at Bartica, British Guiana, Nov. 1888, and deposited in the herbarium of the Royal Botanic Gardens at Kew. *S. brevifolius* is related to *S. anomalus* Ruhl., but differs in its much shorter leaves, with firm texture and prominent nerves, and its shorter, glabrous, truncate peduncular sheaths with obtuse lamina.

Syngonanthus longipes n. sp. Caudex short and stout, woolly; basal leaves densely cespitose, firm or rigid, ensiform-linear, 6–9 cm. long, 3–6 mm. wide, obtuse, glabrous, the nerves about 9, prominent beneath; flowering branch single, 2–3 dm. long, terete, glabrous; involucral leaves resembling the basal, numerous, acute; peduncles 4–14, each 2–4 dm. long, somewhat twisted, costate, glabrous, their sheaths obliquely cleft, multistriate, acute; heads subglobose, 4–6 mm. wide; outermost bracts nearly rotund, the middle ones broadly obovate, the inner oblong-obovate, all glabrous, rounded at apex, ranging from 1.7 mm. long and 1.4 mm. wide to 2.3 mm. long by 1.2 mm. wide; subtending bracts narrowly obovate-oblong, 2.2 mm. long, just equaling the flowers; staminate flower: sepals obovate, 1.5 mm. long, 0.7 mm. wide, subacute, nearly glabrous; corolla tubular for 0.4 mm., campanulate above; pistillate flower: sepals narrowly obovate-oblong, 2.0 mm. long by 0.8 mm. (posterior) or 0.6 mm. (lateral) wide, obtuse, scarious at margin, nearly glabrous; petals 2 mm. long, densely hirsute, especially below, clearly separate at base, 0.4 mm. wide at the middle, connate above into a subulate, claw-like organ beyond the styles; ovary 0.7 mm. long; style, including appendages, 0.5 mm. long; stigmas filiform, about 1.0 mm. long.

Type, *Appun 1199*, from the Roraima district of British Guiana, in the herbarium of the Royal Botanic Gardens at Kew; other specimens are *Schomburgk 1060* and *Im Thurn 33*. The type was reported by Oliver as *Paepalanthus Schomburkgii* Klotzsch and this name is written on the Appun sheet but not on either of the two Schomburgk specimens. The descriptions of *P. Schomburgkii* do not agree with this plant in many important respects, nor do they cite any collection number.

Syngonanthus tricostatus n. sp. Leaves densely cespitose on a short caudex, ensiform-linear, thick, rigid, subcartilaginous with obtuse margins, 4–7 cm. long, 2–3 mm. wide, sharply acute, strongly twisted, many-nerved, glabrous, gradually dilated at base; peduncles solitary, 20–25 cm. high, strongly twisted, tricostate, glabrous, the sheath 35–65 mm. long, about equaling the leaves, oblique, sharply acute and subscarious at the apex; heads campanulate, 6–7 mm. high, the involucre 4 mm. high, conspicuously exceeded by the silvery flowers; bracts regularly imbricate, glabrous, broadly ovate-oblong, the exposed portion rotund, brown, obtuse or rounded, scarious at the margin; receptacle flat; staminate flowers: pedicels 2 mm. long; sepals separate, erect, scarious, narrowly oblong-elliptic, 2.7 mm. long by 0.5–0.6 mm. wide, glabrous, obtuse or subacute, the lateral slightly falcate; petals connate below for 1 mm. into a short, slightly funnelform tube, thence separate, erect, narrowly oblong-ovate, obtuse or subacute, scarious, glabrous, the free lobes 3 mm. long, 0.8 mm. wide; filaments filiform, anthers 2-thecous; pistillate flowers: pedicels none; lateral sepals narrowly oblong, conspicuously falcate, 2.2 mm. long, acute; posterior sepal 2.6 mm. long, symmetrical, narrowly oblong-lanceolate, acute; all sepals hyaline and glabrous; petals 3.5 mm. long, strictly erect and connivent, narrowly spatulate, the hyaline glabrous blade closely involute and appearing subulate, lower fourth of the petal densely hirsute with erect brown hairs covering the whole claw; ovary sharply 3-angled.

Type, *R. H. Schomburgk*, first collection, without number, from the Roraima district in British Guiana, deposited in the herbarium of the Royal Botanic Gardens at Kew. If differs from *S. xeranthemoides* in its very thick, glabrous, subcartilaginous, obtuse-margined, strongly twisted, proportionately broader, actually shorter leaves, the longer peduncular sheaths, the smaller heads and the more scarious-margined bracts.

Abolboda rigida n. sp. Leaves cespitose, numerous, erect, 15–20 cm. long, 5–7 mm. wide, thick and rigid, prominently

dilated at base, pungent at apex; scapes 3, rather stout, glabrous, 6–8 dm. high; bracts a single opposite pair somewhat above the middle of the scape, loosely erect, broadly linear, sheathing and overlapping below, scarious-margined, tapering to the pungent tip, 3–5 cm. long; heads broadly ovoid or subglobose, 14–18 mm. long, many-flowered; involucral bracts firm, scarious at margin, finely veined, the outermost ovate, the inner gradually elongate; sepals 2, naviculate, carinate, oblong, 13 mm. long by 3 mm. wide as folded; corolla-tube slender, 13 mm. long, its lobes broadly round-obovate, 10 mm. long; filaments slender, nearly 5 mm. long; anthers 2 mm. long, prominently prolonged at base; ovary obovoid, 4 mm. long; style very slender, 15 mm. long, the two slender appendages attached 2 mm. from the base, 4 mm. long and abruptly recurved near the middle.

Type, *Schomburgk 146.5*, from British Guiana, in the herbarium of the Royal Botanic Gardens at Kew. It is most closely related to *A. poarchon* Seub., of which I have seen the type. The latter is a smaller, more slender plant with shorter and narrower leaves, somewhat narrower and more pointed involucral bracts, and narrower, conspicuously acuminate sepals.

Xyris cyperoides n. sp. Section *Nematopus;* cespitose, essentially acaulescent, the leaves at base more or less surrounding the scapes and often somewhat twisted and stemlike in aspect; leaf-blades filiform, 10–16 cm. long, 0.3–0.4 mm. wide by half as thick, the edges obtuse, the sides minutely scabrellate under a strong lens, gradually expanded below into a narrow scarious sheath; scapes about equaling the leaves, not flattened, several-ribbed; heads obovoid, becoming obconic under pressure, 3–4 mm. long; bracts erect or loosely appressed, becoming spreading when pressed, obscurely carinate, the outer most ovate-lanceolate, acute, the upper narrowly ovate, acuminate, only the uppermost one or two fertile; lateral sepals free, 3.5 mm. long, linear-spatulate, carinate, inequilateral, the narrower side firm and entire, the broader side hyaline and often lacerate near the middle, the keel entire; petals and stamens not seen; placentae basal.

Type, *Jenman 1056*, from Kaieteur savanna, British Guiana, Sep.–Oct., 1881, deposited in the herbarium of the Royal Botanic Gardens at Kew. It is a member of Malme's *Stirps X. guianensis*, and is most closely related to *X. subuniflora* Malme and *X. Grisebachii* Malme. From the former it differs in its long leaves and narrow sheaths.

Rapatea viscosa n. sp. Leaves all basal, linear, 2 m. long, the basal sheath flat, 3 dm. long, 25 mm. wide, the blade 4 cm. wide, narrowed to the base; peduncles stout, erect, 6–9 dm. high, strongly angled and dilated above; involucre of four bracts, at first erect, more or less connate, enclosing the heads, and covered with mucus, later separate and reflexed, brown, membranous or subscarious, the outer pair narrowly triangular, 10 cm. long, 30–35 mm. wide at the base, the inner linear-triangular, 4–7 cm. long, 1 cm. wide at base; heads semiglobose, 4 cm. in diameter, on a broadly conic, fleshy receptacle 15 mm. high; flowers sessile; bracts about 15, stiff, coriaceous, imbricate, ovate-lanceolate, acuminate to a short subulate tip, the largest 14 mm. long, the innermost slightly shorter; sepals lanceolate, bracteoid, long-acuminate, 12 mm. long.

Type, *Altson 388*, common among sandstone boulders, on top of the escarpment, Kurupung Mountains, British Guiana, 1 Sep. 1925, deposited in the herbarium of the Royal Botanic Gardens at Kew. If differs from the other three Guiana species in the presence of four bracts enclosing the sessile flowers.

Stegolepis angustata n. sp. Acaulescent herb; leaf-sheaths distichously imbricated at base, strongly folded longitudinally, firm or subcoriaceous, narrowly elliptic, 2–5 cm. wide, 15–20 cm. long, gradually narrowed to the acute apex, finely nerved; leaf-blades linear, 2–3 cm. wide, 6–8 dm. long, gradually acuminate to an obtuse tip, long-cuneate at base, finely many-nerved; scape axillary, apparently single, 8–12 dm. tall, slender, prominently angled below, somewhat quadrangular and flattened above; involucre none; heads 3–4 cm. in diameter, the flowers sessile, 15–17mm. long; bracts pale brown, coriaceous, rigid, regularly imbricate in several series, obtuse, the outer short-ovate, the inner oblong; sepals ovate-lanceolate, acute, 15 mm. long; petals with sub-rotund blade, the claw-like bases somewhat connate; anthers linear-lanceolate, 4-locular, opening by a single oblique terminal pore; ovary 3-celled, the ovules several in each cell.

Type, *Jenman 958*, collected on the Kaieteur savanna, British Guiana, Sep.–Oct. 1881, and deposited in the herbarium of the Royal Botanic Gardens at Kew. It differs from *S. guianensis* Klotzsch, of Roraima, in its tapering sheaths, cuneate leaf-bases, and pale brown bracts; from *S. ferruginea* Baker, also of the Kaieteur region, in its tapering sheaths, pale brown bracts, solitary peduncles, and large flowers and heads.

Tofieldia rupestris n. sp. Leaves cespitose (not distichous), coriaceous, at the summit of a short stout rhizome, broadly

linear, erect and spreading, 4–9 mm. wide, 6–15 cm. long, long-acuminate, glabrous; scape 1–2 dm. high, the upper half or two-thirds floriferous; peduncle with several appressed, scarious, linear bracts 1–2 cm. long; pedicels single, subtended by scarious lanceolate bractlets 3 mm. long; pedicels 5 mm. long at anthesis, spreading, becoming 1 cm. long in fruit; involucel none; flowers perfect, hypogynous; perianth-segments distinct, white, petaloid, thinly membraneous, obtuse, in two broadly overlapping series, 3 mm. long, the outer ovate, 1.2 mm. wide, 3-nerved, the inner elliptic, 1 mm. wide, 1-nerved; stamens 6, erect, distinct, inserted on the base of the perianth; filaments linear-oblong, flat, 1.5 mm. long; anthers 2-celled, linear-oblong, 1.1 mm. long; ovary slightly 3-lobed, conic, 2.5 mm. long, tapering regularly to the acute summit, 3-celled with about 8 ovules in each cell; styles 3, separate to the base, very short, with 3 capitate stigmas, the total length of both 0.4–0.5 mm.; capsule septicidal; seeds oblong.

Type, *Altson, 326*, collected from moist sandy crevices on boulders, in shade, Macreba Falls, Kurupung River, British Guiana, 28 Aug. 1925, and deposited in the herbarium of the Royal Botanic Gardens at Kew. *Altson 393*, collected a week later from the same place, exhibits mature capsules.

Smilax latipes n. sp. Stems climbing, the upper branches nearly free from prickles; petioles stout, twisted, 10–12 mm. long; stipules ovate-oblong, 5–6 mm. long, not usually cirrhiferous; leaf-blades firm, ovate-oblong, 12–18 cm. long, 4–8 cm. wide, acute, rounded at base, 3-nerved, with an obscure outer pair additional; veinlets prominently reticulate; flowering branches 15–30 cm. long, their leaves resembling the cauline but thinner and smaller; peduncles stout, flattened, 2–3 cm. long; umbels many-flowered; pedicels slender, 1 cm. long; segments of the staminate perianth spreading, oblong to oblong-elliptic, 4 mm. long, the outer series 1.5 mm., the inner 1.2 mm. wide; filaments flattened, 1.0 mm. long, 0.7 mm. wide at base, half as wide at the summit; anthers ovoid, acute, straight, 0.8 mm. long.

Type, *Gleason 294* (staminate flowers only), collected at Tumatumari, British Guiana, 18 June–8 July, 1921, and deposited in the herbarium of the New York Botanical Garden. Two other specimens in the herbarium at Kew, *Schomburgk 220* (*159*) and *Jenman 4050*, are also referred here. *S. latipes* is very similar in habit to *S. Schomburgkiana* Kunth, but differs in its much larger flowers and proportionately broader filaments.

Costus bracteatus n. sp. Stem stout, glabrous, height not stated, at least 1 cm. in diameter below; leaf-sheaths very finely scabrellate; ligules prolonged 6–10 mm. above the base of the petiole and 10–20 mm. on the opposite side, depending on the size of the leaf, finely veined, minutely pubescent, sparsely hirsute-ciliate; petiole differentiated, 4–10 mm. long, finely pubescent; leaf-blades oblanceolate, 33–39 cm. long, 9–9.5 cm. wide, the uppermost smaller, down to 14 by 4 cm., sharply acuminate, tapering from above the middle to the narrow but obtuse base, glabrous above, very minutely pubescent beneath or almost glabrous in the middle and upper parts; peduncles 24–30 cm. long, 11–12 mm. in diameter, strongly striate, conspicuously but finely pubescent; peduncular sheaths slightly shorter to slightly longer than the internodes, in one-third phyllotaxy, cylindric, not at all spreading, 4–5 cm. long on one side, 5–6 cm. long on the other, somewhat retuse at the summit on the shorter side, finely scabrellate to the touch, minutely pubescent under the lens, varying to nearly glabrous toward the base or perceptibly pubescent distally; spikes stoutly ovoid, 6–7 cm. long, 4 cm. in diameter, densely flowered, the rhachis half as long as the spike; bracts membranous, broadly ovate, appressed, 3–4 cm. long, essentially glabrous, their appendages recurved or spreading, ovate to elliptic, membranous, the lower 8 cm. long by 5 cm. wide, the upper reduced to ovate tips 1 cm. long and wide, veiny and minutely pubescent.

Type, *Jenman 6504*, collected on the Mazaruni River, March, 1892, and deposited in the Jenman Herbarium; a second collection is *Jenman 6818* in the same herbarium, collected in British Guiana without definite locality and labeled "perianth yellow, within finely veined in crimson." The species differs greatly from all others in the region in its long peduncle covered with leafless sheaths. The relation and grouping of the species depends on the location of the peduncle and this can not be determined positively from the specimens at hand. Apparently it is basal, since all species with terminal spikes, so far as known to me, are leafy to the spike, the upper portion of the stem is much smaller in diameter than the peduncle, and our plant bears a general habital similarity to *C. geothyrsus* K. Schum., in which the peduncle is definitely basal. If this interpretation is correct, *C. bracteatus* is the third known American species with this habit and differs from the Ecuadorean *C. geothyrsus* and the Peruvian *C. erythrocoryne* K. Schum. in the presence of terminal appendages on the membranous bracts.

Calathea fragilis n. sp. Herbaceous from a woody rhizome; leaves all basal, 2–6 in number; petioles incomplete, the upper portion flattened and grooved or deeply striate to the geniculus; geniculus smooth, almost terete, 7–9 cm. long; blades narrowly ovate, 61–63 cm. long, 29–31 cm. wide, rounded or subcordate at base but cuneate at the insertion of the petiole, convexly narrowed to an obtuse but apiculate apex, the apiculum 5–10 mm. long, glabrous; peduncles terete, glabrous below, becoming subtomentose distally, with one or two convolute basal sheaths as much as 2 dm. long and sometimes with a single, convolute, linear-elliptic, long-acuminate sheath 25 mm. wide and 20 cm. long about 15 cm. below the head; heads ovoid, appearing globose when pressed, 8.5–11 cm. long, densely flowered; bracts soft, membranous or subscarious, loosely appressed, unappendaged, finely longitudinally veined with numerous cross veinlets, the tissue very delicate or evanescent; outermost bracts broadly ovate, sessile and clasping, obtuse or subacute, 5–7 cm. long, 3–5 cm. wide, densely short-tomentose on the outside near the base, the remainder glabrous; upper bracts spirally imbricate, rotund, 4–4.5 cm. long and a little wider, the uppermost reduced to 2.5–3 cm. in length and width; bractlets erect, stiff, 22–26 mm. long, acuminate, with numerous longitudinal veins, fragile and fissile; peduncles 6 mm. long; bractlet (prophyllum) linear-subulate, triquetrous, slightly clavellate, 18 mm. long; sepals separate, connivent, lanceolate, 17 mm. long, 4 mm. wide, longitudinally veined, fragile; corolla-tube very slender, 18 mm. long; petals 3, elliptic, 15 mm. long, 5 mm. wide, acuminate, equal; exterior staminode broadly obovate-rotund, 12 mm. long, 9 mm. wide; cucullate staminode erect, oblong, 13 mm. long, with a recurved lateral acute process 2 mm. long; style stout, triquetrous, curved at the apex into a complete circle.

Type, *Altson 495*, collected at Erengtaru, Maicwac River, British Guiana, in forest on hill slope in red loam, alt. 1800 ft., April, 1926, and deposited in the herbarium of the New York Botanical Garden; name in Patamona dialect *Turura*; with the notes by the collector 'A tall herb. Leaves arising from a woody rhizome, 2–6 in a clump. Petioles 6 ft. long, blades 2 ft. long, 14 in. wide. Peduncle 20 in. long. Inflorescence ovoid, groups of (?) 5 flowers subtended by a chaffy bract. Flowers cream-colored. Fruit orange, trigonal, fleshy, 3-celled. Seed hard.'

Our plant clearly belongs in Petersen's section Nudiscapae, among the four other species with subulate bractlets, numbers 47–50 of Schumann's monograph. Of these, *C. altissima* (Poepp.

and Endl.) Koern. is most closely related, having also delicate fissile bracts, but differing in its narrower, sharply acute bracts more thinly pubescent, its shorter pubescent sepals, and larger corollas and staminodes.

Gymnosiphon breviflorum n. sp. Stems slender, simple, erect, 1–2 dm. high; leaves reduced to scattered elliptic scales 1 mm. long and 1–3 cm. apart; racemes geminate, separated by a terminal flower, 1–5 cm. long, 3–10-flowered; bracts elliptic, minute, obtuse; pedicels becoming 4–5 mm. long, or the uppermost a trifle shorter; perianth-tube slender, 4 mm. long, its three lobes ovate, 3 mm. long, soon dehiscent, the persistent base 6-nerved, becoming stoutly cylindric.

Type, *Hitchcock 17,349*, growing among leaves on the forest-floor, Tumatumari, British Guiana, 3–5 Jan. 1920, and conserved in the herbarium of the New York Botanical Garden; also *Bartlett* without number from Kangaruma and *Jenman 7191*, in part, from the Mazaruni River.

Gymnosiphon guianense n. sp. Stems slender, simple, erect, 10–25 cm. high; leaves reduced to scattered elliptic scales 1 mm. long and 1–3 cm. apart; racemes geminate, separated by a terminal flower, 2–10 cm. long, 5–10-flowered; bracts minute, ovate, obtuse; pedicels rarely exceeding 2 mm. in length; perianth-tube 5–6 mm. long, tapering above, its three lobes ovate, 3 mm. long, soon deciduous, the tube truncate-conic, becoming 6 mm. long, conspicuously nerved above and subulate along the side of the ovary.

Type, *Im Thurn* without number, collected at Tukeit, Potaro River, British Guiana, 22 Feb. 1879, and deposited in the herbarium of the Royal Botanic Gardens at Kew. It is also represented by *Jenman 7191*, in part, *Lang 131*, and *Altson 304* from the Mazaruni River, *Abraham 216* from the upper Berbice River, *Bartlett* without number from Onderneeming, *Appun 686* and *Jenman 1763* without definite locality.

All the species of *Gymnosiphon* hitherto known from north-eastern South America were described by Bentham in 1855 under the genus *Ptychomeria* and all the types may be seen in the herbarium at Kew. No species have been described since then and no attempt has been made at Kew to assign specific names to the Guiana material. Bentham's species differ markedly from the two described above in their smaller size, smaller and usually slender perianth, and reduced or capitate

inflorescence. Our two species, one of which appears to be widely distributed, may be distinguished at a glance by the combination of long pedicels and short flowers in *G. breviflorum* and short pedicels with long flowers in *G. guianense*.

Gymnosiphon Altsoni n. sp. Saprophytic; stem erect, simple, slender, from a single long slender root, 5–8 cm. high, beset at intervals of 6–8 mm. with lance-subulate appressed scales 2–3 mm. long; inflorescence a simple raceme of 1–3 flowers; bracts ascending, lanceolate, 3–4 mm. long; pedicels 1–2 mm. long; perianth-tube, including the ovary, 7–8 mm. long, 6-ribbed, slender, tapering toward the apex; perianth-lobes 3, ovate-lanceolate, spreading, 5 mm. long, white, yellow in the throat; filaments 0.6 mm. long, inserted in the throat; anthers 2-celled, 0.5 mm. long and wide, each theca somewhat constricted in the middle; style slender; stigmas 3, flattened, obcordate; corolla-limb circumscissile after anthesis just above the insertion of the filaments; ovary 1-celled with parietal placentae; ovules subglobose; capsule and seeds not seen.

Type, *Altson 364*, on the floor of the forest in sandy soil, Macreba Falls, Kurupung River, British Guiana, 30 Aug. 1925, deposited in the herbarium of the Royal Botanic Gardens at Kew. *Altson 305*, from the same locality, is identical. The collector notes that the stem is violet, the flower-buds brown-lilac, and the stigma violet. The species is included in *Gymnosiphon* with some reluctance. The plant has the aspect of *Campylosiphon* Benth., but its 1-celled ovary and wingless filaments place it in or near *Gymnosiphon* or *Dictyostega*. It resembles the former of these in the shape of the 3-lobed perianth and probably also in the seeds, provided that they maintain the same shape as the ovules. The known species of *Gymnosiphon*, however, have flowers in geminate clusters, and the corolla falls below the insertion of the filaments rather than above.

Some Juncus names

KENNETH K. MACKENZIE

I. CONCERNING JUNCUS TENUIS

Juncus tenuis Willd. (Sp. Pl. **2**: 214. 1799), 'Habitat in America boreali,' is *Juncus dichotomus* Elliott (Bot. S. Carolina & Georgia **1**: 406. 1817), and is not the plant appearing as *Juncus tenuis* in our current manuals. Dr. Diels, Director of the Botanical Garden and Museum at Berlin, has very kindly sent me from Willdenow's material portions showing the leaf-blade, the mouth of the sheath, the inflorescence, and the seeds. The leaf-blade is not flattened, and the auricles are rounded cartilaginous and not conspicuously prolonged.

I was brought to look into this matter by noting that Steudel (Syn. Pl. Glum. **2**: 305. 1855) described *Juncus tenuis* Willd. as with 'vaginis adpressis ore nudis.' On the same page he described a new species *Juncus germanorum* ('*J. tenuis* Auctor. Germ.') as with 'vaginis laxiusculis ore in marginem membranaceum utrinque productis (ligulam mentientem).' This last, of course, is a very accurate description of the plant appearing in our manuals as *Juncus tenuis*.

Our current treatments of the *Juncus tenuis* group follow articles by Wiegand (Bull. Torrey Club **27**: 511–527. 1900; **30**: 446–448. 1903). From a systematic standpoint these articles are most admirable, and bring out the specific lines well, largely by the use of sheath characters. But unfortunately Wiegand made no attempt at all to look into the synonymy. Had he studied the work of Steudel he would not have made the statement that 'the characters used, it will be seen, are in many cases quite different from those commonly employed for the separation of species of Juncus.' In fact Elliott (*l.c.*) had used them as early as 1817, when he described what he took for *Juncus tenuis* as having 'stipules membranaceus, 1–2 lines long, bifid.'

Buchenau (Engler, Pflanzenreich 4^{36}: 115–120. 1906) followed Wiegand closely. His descriptions and illustrations are of a very high character indeed. In fact they are among the

best and most helpful in any group. Unfortunately he too does not seem to have always checked up the work of older authors.

Looking through the Index Kewensis and also through the synonymy given by Buchenau one finds a number of names given as synonyms of *Juncus tenuis* Willd. I have investigated the earlier of these names as follows:

(1) *Juncus bicornis* Michx. Fl. Bor.-Am. **1**: 191. 1803. This species came from Georgia and Carolina. It is described as with 'foliis radicalibus, subsetaceis, canaliculato-planis' and 'involucro setaceo-2-phyllo erecto, paniculam terminalem parvulam superante.' It seems to me that Elliott (Bot. S. Car. & Ga. **1**: 406. 1817), who more fully and very definitely described this species, did so correctly, and that the name *Juncus bicornis* Michx. is the name we should use for the species appearing in our manuals as *Juncus tenuis* Willd. Michaux referred to plates by Rostkovius (Monogr. Junc. 24. *pl. 1. f. 3.* 1801) and by Morison (Hist. Pl. **3**: 228. Sect. 8. *pl. 9. f. 15.* 1699). Morison's early plate is based on a plant from Virginia 'Gramen junceum Virginianum calyculis paleaceis bicorne.' The illustration is not a good one, but it certainly shows flattened leaf-blades and I believe is quite properly referred here. It seems to me too that the plant illustrated by Rostkovius is also to be referred here or possibly to *Juncus Dudleyi* Wiegand.

The early illustration of Gramen junceum elatius, pericarpiis ovatis Americanum by Plukenet (Alm. 179. *pl. 92. f. 9.* 1696) also belongs here, I believe. As far as I know this is the earliest plate of an American species of this group. An extraordinary thing is that Linnaeus cited both the above referred to plates of Morison and Plukenet in his original description of *Juncus nodosus* (Sp. Pl. ed. 2. **1**: 466)—a species with which they have nothing in common. This was long ago noted by Rostkovius (*l.c.* 25).

A photograph of part of the original material of *Juncus bicornis* from 'Georgie' in Herb. Mus. Paris has been kindly sent me by Professor Lecompte. It bears Dr. Engelmann's identification '*Juncus tenuis* Willd. G. Engelmann 1869.' This particular specimen, however, does not answer the description of Michaux. The bracts are shorter than the inflorescence and the leaf-blades do not appear at all flattened. The auricles, too,

are apparently short and not prolonged. However, in view of the description in Michaux, it seems to me that we should use his name as did Elliott for the plant we have been calling *Juncus tenuis* Willd. Michaux had material from both Georgia and Carolina, and one is left with the impression that his description was not based on the Georgia material.

(2) *Juncus gracilis* J. E. Smith (Engl. Bot. 31. *pl. 2174.* 1810; not *J. gracilis* Roth Tent. Fl. Germ. **1**: 155. 1788; not *J. gracilis* R. Br. Prodr. 259. 1810) seems to be the next name to be considered. This was based on specimens collected in Angusshire, Scotland. (The species occurs widely in Europe, where it is regarded as introduced.)The illustration of the leaf sheaths is of a conventional character, but as Smith (Eng. Fl. **2**: 167. 1824) described the leaves as dilated and membranous at the base, I would regard this species as the same as *Juncus bicornis* Michx. as here treated.

Smith's name being a homonym, three other names were proposed by different botanists, each name being strictly synonymous with his. These names are *Juncus macer* S. F. Gray (Nat. Arr. Br. Pl. **2**: 164. 1821); *Juncus Gesneri* J. E. Smith (Eng. Fl. **2**: 167. 1824); and *Juncus Smithii* Kunth (Enum. Pl. **3**: 349. 1841). The first of these is apparently the next name available for the '*Juncus tenuis* Willd.' of our floras, should *Juncus bicornis* Michx. turn out not to be available.

(3) *Juncus platycaulos* H.B.K. Nov. Gen. et Spec. Pl. **1**: 190. (236 in smaller ed.) 1815. This was based on South American material collected 'in ripa Orinoci prope Atures et Caricharia.' According to Buchenau (Pflanzenreich 4^{36}: 117) this does not possess the 'auriculas magnas scariosas Junci tenuis' (*J. bicornis* Michx. as here treated), and apparently from a poor specimen it represents *Juncus dichotomus* Ell. It will be noted that its publication antedates that of *Juncus dichotomus* Ell.

II. CONCERNING JUNCUS SETACEUS ROSTK.

Juncus setaceus Rostk. (Mon. Junc. 13. 1801) is a species which has been most persistently misunderstood. It is based on an immature plant collected in Pennsylvania ('Habitat in Pennsylvania'), and is described as leafless ('J. culmo nudo'). It is said to be 'affinis valde praecedenti,' the immediately

preceding species being *Juncus filiformis* L. The number of stamens is not given. The perianth divisions are described 'petalis subulatis,' and 'apice subulato' and are figured with tips such as are very noticeable in young *Juncus effusus*, but not in young specimens of the plant we have been calling *Juncus setaceus*.

Rostkovius gives an illustration of his species (*pl. 1. f. 2*), which shows the upper part only of an immature plant.

It has been identified with a plant which has culms leafy at the base, and which is especially noticeable for its stiff,rigid, soon spreading perianth parts. This last mentioned plant is a coastal plain plant, reaching its northern limit in Delaware and the southern part of Cape May County, New Jersey.[1]

While then neither the description given by Rostkovius nor his type locality fit the plant which has been passing under the name *Juncus setaceus*, yet description, type locality, and plate do fit the common Pennsylvania form of *Juncus effusus* L. Accordingly I am treating *Juncus setaceus* Rostk. as a synonym of *Juncus effusus* L. Engelmann (Trans. Acad. Sci. St. Louis **2**: 444–5. 1866; Botanical Works 249) was much troubled about the identification of this species, but did not follow up the matter.

For the plant which appears in our manuals as *Juncus setaceus* Rostk. (Brit. & Brown, Ill. Fl. ed. 2. **1**: 473. *f. 1184;* Small, Flora S.E. U. S. 258; Gray, Man. ed. 7. 272. *f. 575;* Buchenau in Engler, Pflanzenreich **4**[36]: 123) I propose the name **Juncus coriaceus**, referring to the thick rigid texture of the sepals, petals and capsules. As a type may be designated a specimen in my herbarium collected by Henry Eggert, July 9, 1896, at Hot Springs, Arkansas, in swamps.

[1] In Tatnall's Catalogue of Plants of Newcastle County, Delaware (1860) p. 80, it is given as follows: 'Wet grounds; Appoquinimink. Rare.' In Cape May County, New Jersey it is 'of frequent occurrence, though local' (Stone, Plants of Southern New Jersey 332. 1911).

It is reported from Tinicum, Delaware county, Pennsylvania (Dr. George Smith) in Keller and Brown, Flora of Philadelphia (1915) p. 95, on the authority of B. H. Smith, and apparently this same record appears in Porter's Flora of Pennysylvania, p. 79. This material I am informed represents some species of the *Juncus tenuis* group.

III. CONCERNING JUNCUS CANADENSIS J. GAY

In Rhodora **6**: 35–6. 1904 Professor M. L. Fernald well and carefully pointed out the distinctions between the stout plant with many-flowered heads, which he treated as *Juncus canadensis* J. Gay, and the more slender plant with few-flowered heads which he treated as *Juncus brevicaudatus* (Engelm.) Fernald. The former plant he stated is a plant 'commonest on the coastal plain from southern Newfoundland to Louisiana, and on the upper St. Lawrence and the Great Lakes, though it is occasionally found at other inland stations,' while the second plant 'abounds . . . . from Newfoundland to the upper Saguenay, west to Minnesota, and south, mostly in cold bogs, to the mountains of Pennsylvania.' To this it may be added that the plant first referred to is found in acid soils, while the second is a strong calciphile.

Buchenau (Engler, Pflanzenreich **4**[36]: 160. 1906) objected to the application by Fernald of the name '*brevicaudatus*' to a plant with long-tailed seeds, and in its place took up the name '*J. coarctatus* (Engelm.) Buchenau.'

In looking this matter up, naturally the first question was to find out what *Juncus canadensis* J. Gay was, and whether the name should be applied to a very common and abundant Canadian plant, or to one local and uncommon there. The original description (in La Harpe, Monogr. Junc. in Mem. Soc. Hist. Nat. Paris **3**: 134. 1827 [reprint p. 46]) calls for a plant

> Spicis 4-8-16-floris . . . . ; perigonii foliolis . . . . capsula oblonga prismatica brevioribus. . . . . culmus 6–12 poll. altus . . . . panicula composita, erecta, plus minusve coarctata vel laxiuscula . . . . perigonii foliola lanceolato-linearia, substriata, acute aristata, interiora sublongiora. . . . . capsula ellipsoideo-prismatica, triquetra, utrinque leviter attenuata, vix mucronata.

It was divided into two varieties

> α. Culmo foliisque tenuibus, panicula sublaxa, spicis 10–15-floris remotiusculis.
>
> β. Culmo foliisque tenuibus, panicula coarctata, spicis 4–8- floris approximatis.

Specimens were cited 'Habite le Canada (herb. Michx.), la baie d'Hudson (Torrey), Terre-Neuve (La Pylaie).'

In our current manuals the name has been applied to a plant correctly described in Gray's Manual (ed. 7) as having

'stems stout . . . . 5–50-flowered heads . . . . sepals and petals awn-pointed, mostly shorter than the abruptly short-pointed capsule' (p. 274) and in the key (p. 269) 'capsule equaling or slightly exceeding the calyx.'

On the other hand *Juncus brevicaudatus* is there correctly described 'stem slender . . . . . 3–7-flowered heads . . . . sepals acute, the petals rarely obtusish, much shorter than the prismatic gradually pointed deep-brown capsule' (*l.c.* 274) and in the key 'capsule much exceeding the calyx gradually tapering' (*l.c.* 269). It is very beautifully illustrated by Buchenau (*l.c.* 160).

Investigation of original material showed the following:

(1) A photograph of the Canadian material in Museum d'Histoire Naturelle at Paris collected by Michaux shows on one sheet one plant of *Juncus brevicaudatus* (Engelm.) Fernald and two small plants of another species. This sheet bears Dr. Engelmann's identification made in 1869 that the first mentioned specimen is '*Juncus canadensis* var. *coarctatus*' and the small plant is '*Juncus alpinus*, forma *pallida* depauperate.' The original label bears the wording 'Baye d'Hudson,' and shows that this locality attributed to Torrey by Gay really belongs to the immediately preceding citation from Michaux.

(2) Two photographs of the Newfoundland material of La Pylaie have been received. One from Museum d'Histoire Naturelle at Paris is of a sheet containing seven specimens. It bears the determination '*J. canadensis* var. *coarctatus* Engelmann 1869.' A sheet in the Kew herbarium contains two plants of La Pylaie material. These agree with the Paris material.

It will be seen from the above that all of the Canadian and Newfoundland material cited by Gay represents his variety β. They all represent the plant now generally called *Juncus brevicaudatus* (Engelm.) Fernald, as marked by Engelmann.

(3) The only material cited by Gay unaccounted for above is the Torrey material, incorrectly given as collected 'la baie d'Hudson.' Investigation at Paris did not result in finding any such material. A photograph from a specimen in the Hooker herbarium at Kew was received with the following note:

> The right-hand specimens (the only ones photographed) are labeled *Juncus acuminatus* and that half of the herbarium sheet is endorsed in W. J.

Hooker's handwriting 'Dr. Torrey. *J. Canadensis* La Harpe.' They are probably not the actual specimens seen by La Harpe, but may possibly be duplicates of the same collecting.

As far as I can tell from the photograph, the specimens (four in number) are specimens of *Juncus acuminatus* Michx.

From the above it will be noted that no material seen by Gay has been found answering to our current interpretation of *Juncus canadensis* J. Gay.

The Canadian material collected by Michaux seems to me to be the type material of Gay's species, and accordingly I am proposing to use the name *Juncus canadensis* J. Gay in place of *Juncus brevicaudatus* (Engelm.) Fernald and *Juncus coarctatus* (Engelm.) Buchenau. In passing it may be noted that Gay's capsule description applies to this plant, and not to either *J. canadensis* of our manuals or to *J. acuminatus*. It reads, as quoted above, 'capsula ellipsoideo-prismatica, triquetra, utrinque leviter attenuata, vix mucronata.'

The only evidence found tends to the belief that *Juncus canadensis* var. α. J. Gay was based on specimens of *Juncus acuminatus* Michx., but in view of the non-conclusive character of this evidence this conclusion must be regarded as tentative only.

For the plant which has been appearing in our manuals as *Juncus canadensis* J. Gay I am proposing to use the appropriate name **Juncus longicaudatus** (Engelm.) n. comb. (*Juncus canadensis* var. *longicaudatus* Engelm. Trans. St. Louis Acad. Sci. **2**: 436. 1866 and **2**: 474. 1868).

I am under obligations to Prof. H. Lecomte of the Museum d'Histoire Naturelle at Paris and to Dr. Arthur W. Hill, director of the Kew Gardens, for photographs of the material here referred to.

IV. WHAT IS JUNCUS BREVICAUDATUS (ENGELM.) FERNALD?

Juncus brevicaudatus (Engelm.) Fernald goes back to the following few words

J. Canadensis Gay
α. *brevicaudatus* (J. acuminatus Gray)
A. *coarctatus*. Pennsylvania, northward and north-westward.
B. *patulus*. Pennsylvania to western New York and Ohio.

(Trans. Acad. Sci. St. Louis **2**: 436. 1866; Engelmann's Botanical Works 245.)

The name was quickly abandoned by Engelmann (Trans. Acad. Sci. St. Louis **2**: 474. 1868; Engelmann's Botanical works 263) as Buchenau (Pflanzenreich 4^{36}: 160) points out.

However, Buchenau is in error in stating that the name was *nomen nudum*, because Engelmann did cite Gray's '*Juncus acuminatus* Michx.' This reference is to Gray's Manual ed. 2. 481. 1856). There the description given is, I believe, mostly if not entirely of what we have been calling *Juncus brevicaudatus*. Under the circumstances the name *J. brevicaudatus* (Engelm.) Fernald, although most inappropriate for a plant with long-tailed seeds, can logically be used as in our current manuals. The other way is to proceed on the belief that Engelmann must have based the name upon some specimen of his *J. Canadensis* α *brevicaudatus* B *patulus* which later became his *J. canadensis* var. β *brachycephalus*. (Trans. Acad. Sci. St. Louis **2**: 474. 1868; Engelmann's Bot. Works 263–4). This is a plant with short-tailed seeds, and one to which Engelmann's first name is very well applicable. Gray's description too of his *Juncus acuminatus* Michx. calls for a plant with 'abruptly acute pod,' leading one to believe that he included *Juncus brachycephalus* (Engelm.) Buchenau in his conception of Michaux's species. If this last course is pursued the name beomes a synonym of *Juncus brachycephalus* (Engelm.) Buchenau Monogr. Junc. 268. 1890. I am inclined to believe that this would be the best course.

The name taken up by Buchenau, *Juncus coarctatus* (Engelm.) Buchenau (*l.c.* 160), goes directly back to *J. canadensis* β J. Gay, and is strictly synonymous with *J. canadensis* J. Gay, as I interpret it.

The use of osmic-impregnation methods in plant cytology

ROBERT H. BOWEN

(WITH PLATE 1)

The use of osmic acid for the purpose of blackening certain kinds of cytoplasmic elements has been a familiar method in animal cytology for many years. This is particularly true of the blackening of certain types of fat-materials, which often occurs upon the addition of osmic acid or mixtures containing it. But a much more important effect, and one concerning the chemistry of which very little is known, is the blackening of various lipoidal structures after prolonged exposure to aqueous solutions of osmic acid. This effect was observed certainly as long ago as 1888 by Prenant, but the real significance of the method was first made known by Kopsch ('02). This worker discovered that by immersing animal tissue in 2 per cent osmic acid for periods of several days (or weeks) it was possible to blacken the so-called Golgi apparatus in a most selective manner. This reaction was further studied by Sjövall ('06), who sought to improve the very poor fixation effected by the action of osmic acid alone, by first fixing the material in formalin and subsequently subjecting it to the osmic treatment. This separation of fixation from the osmication step was further studied by Weigl ('10) and finally perfected by Kolatchev ('16). By these methods, now familiar to all animal cytologists, it is possible to effect a blackening of the Golgi material in all kinds of animal cells. This blackening in animal cells is usually restricted to Golgi material, but under certain conditions, which are not yet very well understood, the mitochondria may be blackened, and occasionally other structures. In fact, by a slight variation in one of the usual methods Hirschler ('25) obtained a sharp blackening of the nuclear membrane. It is to be distinctly understood, however, that such results in animal cells are the exception—the rule is that the Golgi apparatus alone is almost invariably the only structure to show a distinct blackening.[1]

[1] This statement does not apply to Protozoa, which present technical problems all their own.

Some of these osmic methods have now been shown to yield results of truly remarkable excellence, when properly handled. Considering the interest which attaches to certain problems connected with the Golgi apparatus, it is remarkable that for over twenty years after its identification in animal cells, practically no effort was made to examine plant material for its occurrence. This is even more difficult to understand, in view of the extensive and controversial literature which had been accumulating on the side of animal tissues. The obvious thing to undertake was clearly the effect on plant cells of these same osmic methods. This was first seriously tried by Nassonov ('18), who applied the method worked out by Kolatchev to various kinds of plant cells. He observed that the plastid-forming bodies in meristem cells were frequently selectively blackened, and that at other times cytoplasmic networks especially associated with the division figures were demonstrated.

Somewhat later (Bowen, '26), I repeated Nassonov's work, using other methods as well as the one employed by him, and found to my astonishment that in different cells it was possible to obtain a precise blackening of practically any cellular structure with the exception of chromosomes in mitosis. Much the same result was reported almost simultaneously by Guilliermond ('26). There is thus placed in the hands of cytologists a method of technical attack of the greatest interest and value, which permits the study of certain cytoplasmic structures otherwise undemonstrable, as well as providing further means for the study of structures already well known.

In view of the fact that most of the work on plant cells by these methods has been done thus far in zoological laboratories, it has seemed to me that botanical workers might welcome some report of the methods arranged particularly for their own purposes. For this reason the present paper has been prepared. In it I shall give detailed directions for the methods which have been shown to work successfully, together with some analysis of the results obtainable. More detailed reports on specific cytoplasmic structures may be found in two of my recent papers (Bowen '28, '29). The methods of osmic impregnation which have proved useful are primarily those of Kolatchev ('16) and of Weigl ('10). Hirschler ('24) has published a variation of the

Kolatchev method with which I have experimented only a little. In general it yields results much like those of the Kolatchev technique.

GENERAL NOTES

The methods of osmic impregnation require rather more careful manipulation than is called for by most of the orthodox cytological techniques, and certain things are so indispensable that special attention will be called to them in the beginning. I should add, however, that the laborious methods of dehydrating and embedding still common in the botanical laboratory are only a waste of time with osmic technique. Reasonably careful handling such as is adequate for routine work in animal cytology is all that is necessary. The osmic methods are so tedious, so variable, and so often unsuccessful, that it is useless to introduce technical refinements at the present stage of their development.

1. Material for osmic work should be absolutely fresh. Do not use plants which have lingered about the laboratory or greenhouse under unfavorable conditions. Use pieces of material as small as possible. Large root-tips and other massive parts will probably not react successfully unless subdivided.

2. Use osmic solutions, both for fixing fluids and for the osmication process, which are positively fresh—not over one to two weeks old—and which have been kept properly clean and preserved from the action of light.

3. Keep the ingredients of fixing fluids in separate stock solutions, and mix them only just before using.

4. All fixation, and especially all osmication steps, *must positively be conducted in glass-stoppered bottles*. If these are not available do not waste time trying these methods. They will not work with cork-stoppered bottles. The glass-stoppered bottles should not be so large that extensive air-spaces are left above the fixing reagents. The most convenient size for plant work is a bottle containing (when completely filled and the stopper in place) about 4 cc., with a mouth sufficiently wide to permit the easy introduction of a pipette. Such bottles are carried in this country by Eimer and Amend of New York City, from whom I have regularly obtained them. After using a bottle for any purpose, wash it in cleaning fluid, and rinse

it under the tap with scrupulous care, finally rinsing in distilled water before drying. Carry on fixation and osmication in the dark. This is most easily done by keeping the bottles in cardboard boxes with deep covers.

5. Once the material has been put in the fixative, handle it thereafter only with wide-mouth pipettes. The use of forceps is very bad after osmication because the tissue becomes brittle and is easily damaged.

6. After the material is finally washed out in running water after osmication, do not spend more than five or six days at the most in the alcohols before embedding in paraffin. For these steps immediately prior to embedding, it is convenient to carry the material in small shell-vials with cork stoppers. After osmication, this use of cork stoppers does no harm.

THE KOLATCHEV METHOD

The essentials of this method were published in 1916 by Kolatchev. The routine here described follows the procedure used by Nassonov ('18) on plant material, with certain variations that I have found useful.

1. Fix for 24 hours in Champy, as follows:

1 per cent chromic acid	7 cc.
3 per cent potassium bichromate	7 cc.
2 per cent osmic acid	4 cc.

Do not overload the fixing bottles. About 10–15 root-tips not larger than those of *Hyacinthus* can be safely placed in 3 cc. of fixative. The bottles should be occasionally agitated gently to assist in the penetration of the fixative.

2. Transfer to a dish of tap water to remove the excess of fixing fluid, and then wash in running water for 24 hours. This step is important, and the washing out of the chromium compounds must be complete. A convenient method for performing this step with large batches of material has been elsewhere described (Bowen, '27a).

3. Transfer the washed material to a dish of distilled water, and thence to fresh glass-stoppered bottles for osmication. In order to avoid dilution of the osmic acid, I find it convenient first to transfer the material to an empty bottle, draw off all the distilled water which may have been carried over, with a

pipette, and then add the osmic acid. About the same relations should obtain between material and volume of osmic acid as in the case of fixation. Do not be stingy with the osmic solution, it is poor economy. The osmication step may be carried out in several ways. I usually separate the material while in distilled water and proceed to try four variations of osmication, as follows.

I. Place the material in 1% osmic acid solution, and then:
   a. Put the bottles in an incubator at 35°C. for periods of 4 to 9 days.
   b. Put the bottles in an incubator at 40°C. for 8 hours, and then transfer the bottles to an incubator at 35°C. without any change of the osmic solution. Leave in this second incubator for periods such that the total time in osmic acid is from 4 to 9 days, as before.

II. Place the material in 2% osmic acid solution, and then proceed:
   a. As in method I*a*.
   b. As in method I*b*.

I usually carry along all four variations simultaneously. If for one reason or another this is not feasible, probably the best single variation is II*b*. In general this offers the best probability of a successful impregnation, but material varies in its behavior and I have found it up to the present time useful to try all the possibilities. After a longer or shorter period in the incubators, the osmic solution almost invariably begins to blacken. This is of no consequence until it becomes very pronounced, and then it is best to pour off the blackened solution, rinse out with distilled water and add fresh osmic solution; or better, rinse the material in distilled water and transfer to a clean bottle with fresh osmic acid. The temperatures of the incubators need not be absolutely exact, but a range of more than one degree above or below the temperature advised, should be avoided. It is well to take the bottles out of the incubator daily and agitate each one gently to assist in the penetration of the osmic solution.

4. When the preceding osmication step is completed, the material is transferred to running water for 24 hours. This washing is less important than that in step 2, but it is well to see that the osmic solution is removed before proceeding.

The time at which to terminate the osmication is the one difficult variable in this method. There is no way to tell when a successful impregnation of anything whatever has been attained. This is a serious difficulty, because if the osmication period is too short, nothing will be blackened, and if too long, a serious over-blackening occurs which usually ruins the material for critical purposes. One is therefore driven by necessity to a method of trial and error, which is very laborious but which alone can give one any satisfactory idea of the success of his procedure. I proceed as follows. Each day, beginning on about the fourth day of osmication, two or three samples are removed from each bottle, with a wide-mouthed pipette.[2] This is repeated daily up to the eighth or ninth day of incubation, by which time the contents of the bottles will be exhausted. The samples thus obtained are washed and carried along separately. After embedding, section samples near the beginning and end of the osmication period, and at some intermediate point. With a little experience, an inspection of the sections will indicate at once where the best result, if any, has been obtained and one can govern further sectioning operations accordingly. As one becomes acquainted with a particular material, the most probable times for a good impregnation become more definite, but a certain amount of trial by sampling is always advisable, for slight differences in the results of osmication are the rule rather than the exception.

5. The material having been washed as noted in the preceding step, it is then run up and embedded in paraffin. Unlike silver impregnation, osmic blackening is very stable, and no unusual precautions are necessary.

6. Cut sections not over 5 micra thick.

7. When the slides are ready, remove the paraffin in xylol, pass down to absolute alcohol, to insure complete dehydration, and then back to xylol and mount in balsam as usual. For routine work this direct method of mounting is generally sufficient. One may, however, wish to bleach or to counterstain the sections, in which case proceed as described on pages 41 and 42.

[2] Be very careful to use a pipette which can not in any way be contaminated with a chromium compound. It is best to reserve a particular pipette for this step.

THE HIRSCHLER METHOD

This method, although worked out independently by Hirschler ('24), is essentially a variation of Kolatchev's procedure. The steps are practically the same as in the Kolatchev method, with the exception of the osmication stage. It is Hirschler's belief that a prolonged osmication at a lower temperature is better than the rapid osmication as practiced by Kolatchev. Hirschler accordingly osmicates (step 3, page 36) in 2% osmic acid at 20° to 25°C. I have used room temperature, also an incubator at 25°C. Osmication progresses very slowly, and samples may be removed over a long space of time. I have followed the course of events (at 25°C.) at intervals of every other day from the fourth to the thirty-third day of osmication. No results of any particular use were obtained before the sixteenth day, and longer osmication periods are more successful.

THE WEIGL METHOD[3]

The basis of this method was worked out by Weigl in 1910, but the details of its manipulation were never published by its inventor. A number of later workers have experimented with its possibilities, and the following directions represent the variation which I have used on plant material.

1. Fix in corrosive-osmic, the usual proportions being as follows:

1 per cent osmic acid	1 part
Mercuric chloride, saturated solution in 0.75 per cent aqueous sodium chloride	1 part

With some kinds of plant material this fixative blackens within fifteen minutes. In case this occurs, pour off the blackened fluid and add fresh fixative, repeating if necessary at a later time. The length of fixation is an important variable in this method, but it can be arrived at only by trial. I usually fix material for periods of 1, 2 and 4 hours. Shorter or longer periods might be tried, but the ones named are most likely to fall within the period of possibly good fixation. It is also

[3] This method is frequently called by animal cytologists, the Mann-Kopsch method. Since Weigl alone is responsible for the essential steps, it has seemed to me shorter and better to name it after the inventor—the Weigl method.

possible to vary the proportions of osmic acid and mercuric chloride in the fixative, but results on animal tissue do not seem to indicate that much is to be expected from such experiments.

2. After fixation (the batches of material from each fixation period being of course kept separate), wash in running water for 30 minutes. Longer washing will do no harm but is not necessary.

3. Then transfer the material to distilled water and thence to the osmic solution. Osmicate in 2% osmic acid. This may be done at room temperature, but in order to eliminate controllable variables as much as possible, it is best to use an incubator at 25°C. As in the Kolatchev method, the osmication step is subject to wide variations, and the best procedure is to remove samples daily from the third to the eighth day. Handle this part of the technique exactly as described under the Kolatchev method (p. 38).

4. Samples removed from the osmic solution are washed for 24 hours in running water.

5. Then pass up through the alcohols, embed, section and mount, as described under the Kolatchev method (p. 38).

6. If it is desired to bleach or counterstain, see pages 41 and 42.

General result. Sections from material prepared by any of the osmic methods have a characteristic appearance. The structures which may have been successfully impregnated are usually an intense black, but sometimes are only grayish. The remainder of the preparation usually assumes a characteristic yellowish tone, although in exceptional cases the background may be almost colorless. The nucleus is easily distinguished, and chromosomes in division stages can usually be recognized without difficulty. Cell walls may be very sharply delimited, but this is not always the case. Faulty impregnations may show a good deal of granular blackening of the cytoplasm, and from this point the results may grade on down to the impossibly poor. A great deal of experience is necessary in order to judge the probable degree of success of a particular preparation, and no rules can be given. The skillful technician and competent observer will gradually learn the ear-marks of a successful osmic preparation, and until some proficiency in

this matter is attained it is easy to commit serious errors of identification and interpretation.

BLEACHING METHODS

Although the direct mounting of sections from osmicated material is the general rule, it is sometimes useful to clear up the background by bleaching. This process often results in preparations of amazing brilliance, but it is perhaps questionable as to how much the picture is improved for purposes of critical study. Certainly, bleaching will not make first-class preparations out of slides which are not pretty good to begin with. Nevertheless, bleaching is worth trying on occasion and for some methods of counterstaining may be essential. There are three bleaching processes in common use:

The turpentine method. Remove paraffin from the sections in xylol and transfer the slides to double rectified oil of turpentine. Bleaching occurs very slowly. Examine the slides occasionally under the microscope and when the proper point is reached wash the slides thoroughly in xylol and proceed as usual to mounting. The time required by this method will be anywhere from 12 to 48 hours. The action of the turpentine must be checked before it has appreciably attacked the blackening of the particular element one wishes to study.

The hydrogen peroxide method. Run the sections down to water, and bleach in a solution of hydrogen peroxide made by adding 2 drops of the common medicinal solution to 50cc. of distilled water. The action is very rapid and must be controlled with the greatest care. Usually the time required is from 15 to 60 seconds, and even with such short exposure to peroxide, the blackening of the desired elements may be seriously affected. Check the bleaching action by a thorough washing in running water (10 or 15 minutes), and then run up and mount as usual.

The permanganate-oxalic acid method. A variation of this old bleaching process is perhaps the best method at present available. I have used it extensively on plant material and find it a most desirable technique, although a little more complicated than the preceding methods. I should recommend it as the method of choice for bleaching.

1. Run sections down to water.

2. Bleach in 0.1% aqueous potassium permanganate. The sections acquire a brownish tone and the osmic blackening is rapidly attacked. The extraction must be very carefully controlled under the microscope. Usually about 10 to 60 seconds is sufficient time. In some cases, visual examination is very difficult and it is often an advantage to experiment with one's watch as the controlling element. Once the proper time is determined, the remaining slides can be very quickly handled without individual inspection.

3. When the proper point in bleaching is reached, wash *quickly* in distilled water and place at once in 0.1% oxalic acid solution. Here the slides should remain until the brown color acquired in the permanganate has disappeared—about one or two minutes.

4. Then wash at once in running water for ten or fifteen minutes, run up and mount as usual.

The results are very fine, and with a little experience few serious mistakes in differentiation will be made. The permanganate solution goes bad very quickly after using, and fresh solutions should be used each time a batch of slides is to be treated.

COUNTERSTAINING

The value of counterstaining osmic preparations of plant material is something of a question because of the limited work which has thus far been done with these methods. Clearly, counterstaining is usually of little value in routine work and as it almost always involves a great deal of time, it seems scarcely advisable to have recourse to it unless with a definite purpose in mind. As a matter of fact, osmicated tissues are very refractory toward stains of every kind and counterstaining is always difficult. Preparations which are not first-class to begin with, are useless for counterstaining, and it is time wasted to try them. Sometimes fairly good results are obtainable by direct staining of osmicated material,but more often it is better or even necessary to bleach first. If the impregnation is faint, bleaching must perforce be omitted anyhow, but such weak osmications are usually accompanied by a better staining capacity of the material, and bleaching is, therefore, less necessary. One finds, as a matter of fact, that the heavier the osmic

impregnation, the less affinity do the sections have for stains. Conversely, the more complete the bleaching, the better the staining result. Unfortunately just as the tissues begin to take the stain in a really satisfactory manner, the impregnation shows an appreciable extraction, and the bleaching must needs be checked before the tissue is in condition for a first-class stain. The use of a counterstain is thus a matter requiring experienced judgment and this can only be acquired by many trials and errors.

For a general contrast stain, anilin dyes such as eosin, light green or orange G, made up in 95% alcoholic solution may be used. These may give a pleasing color-contrast, but do not add materially to the value of the preparations for critical study. The most valuable stain, indeed the only one of any real worth, is acid fuchsin. This may be employed according to any of the Altmann modifications, but as Nassonov ('18) showed, variations of the Kull procedure are the best. By this method it is possible to stain nuclei and chromosomes, and also the chondriosome-like bodies in the cytoplasm. Of these latter elements, those which are capable of producing plastids are less readily stained than the other type—often called chondriosomes by botanical cytologists. Two ways of applying the Kull method may be used, as follows:

1. Run the sections down to water and bleach or not, as one chooses, by the permanganate-oxalic acid method.

2. Stain in Altmann's acid fuchsin. I flood the slide with stain, and then warm it carefully over a flame until vapors are given off. Cool the slide for 2 or 3 minutes, and repeat the warming process.

3. After a total time of about 5 minutes in the acid fuchsin, wash off the stain completely in distilled water.

4. Differentiate in a 0.5% solution of aurantia in 70% alcohol. The time required is usually less than 1 minute, but no rule can be laid down, and the only safe procedure is to examine the slide under the microscope and check the differentiation at the desired point. When one is doing routine staining on a large batch of similar material, it is often easier to differentiate by the clock, having first obtained the approximate time by trials.

5. When the slide is differentiated to the proper point, rinse

it very quickly in 95% alcohol, then 100% alcohol and transfer finally to xylol. Mount as usual in balsam. The 95% alcohol may be omitted. In any event the dehydration must be effected in the shortest possible time—a few seconds at most.

The preceding method stains the nucleus and chromosomes red, and on some occasions will stain cytoplasmic elements also, provided these have not been blackened by the osmic acid. In general, however, cytoplasmic elements which have not been impregnated can only be stained by the complete Kull technique. This raises a difficulty, since in osmicated material the thionin (or toluidin blue) takes so heavily that a properly balanced differentiation can usually not be obtained. In general therefore, to use the second staining procedure, one must have recourse to bleaching, the usual method for this purpose being the permanganate-oxalic acid technique. The second method for using Kull is, therefore, as follows:

1. After bleaching and thorough washing (5 or 10 minutes) in running water, stain in Altmann's acid fuchsin exactly as in the first method.

2. Rinse off completely in distilled water and flood the slide with a 0.05% aqueous solution of thionin. Stain for about one minute. Sometimes a much shorter time is required, but this must be ascertained by trial and no definite rule of any kind can be given. If the staining time is too long, it will be impossible to remove the blue color sufficiently from the cytoplasmic background before the fuchsin stain is too much extracted.

3. Rinse off again in distilled water, and differentiate in aurantia as in the first method. This will usually require less than 30 seconds. In my own work, I have frequently found 10 seconds a sufficient time.

4. Dehydrate quickly, as in the first method, and mount as usual in balsam.

Unfortunately, acid fuchsin preparations of this type are not very permanent, the stain being lost sometimes in a few months. More favorable examples may last a year or two.

In view of the fact that the more complete the bleaching, the better the stain obtainable with acid fuchsin, it may be possible to salvage preparations in which the impregnation has failed, by completely bleaching and then staining by the usual Kull method. If one's material is limited and preparations of

the chondriosome-like bodies in the cytoplasm are very much desired, this method may be of assistance. In general, however, I think it is not to be recommended as a first-class working procedure.

GENERAL RESULTS

It is in general impossible to predict exactly what will be blackened by these osmic methods in any particular case. Sometimes one type of result is obtained, and in another trial of the same material the result may be different. Or, more probably, in one and the same section—even in adjacent cells—the elements impregnated will be different. This may be very discouraging to some workers who still cling to a faith in specific staining methods, but to one whose interest is in the result rather than the method, osmic impregnations are of extraordinary interest. In some kinds of material it is possible on occasion to get an impregnation of practically all the known structures in the cell. Thus one is enabled to study now one element, now another, in different cells of the same preparation; or one may get various combinations of different elements simultaneously demonstrated in the same cell. One thing only is absolutely essential in the study of osmic impregnations, and that is a trained judgement gained by experience. By repeated trials, by extended study of one's material, and by comparison with preparations made by the more customary methods, it is possible to effect a complete separation and identification of all the blackened elements. With this general caution as to the nature of osmic impregnation in plant cells, some more specific statement of the results may be attempted.

1. These osmic methods were originally designed for the demonstration only of the animal Golgi apparatus, and, as noted in the beginning, they are quite remarkably specific in animal cells. We might, therefore, expect a similar action in plant cells, at least to the extent of demonstrating one element in plant cytoplasm with unusual regularity. Such is actually the result obtained. These methods most frequently and regularly blacken a collection of cytoplasmic bodies to which I have tentatively applied the name of 'osmiophilic platelets.' These are probably a new category of cytoplasmic elements, hitherto unnoticed by botanists because they can,

as a rule, be demonstrated only by osmic impregnation. These have been fully described in another place (Bowen '28), where details as to their structure, etc., may be found. The most common result of osmic impregnation is, therefore, the demonstration of the osmiophilic platelets alone. This has now been effected in bryophytes (fig. 1, and also Bowen '27b), pteridophytes (Bowen '28), gymnosperms (unpublished), and angiosperms (fig. 2, and also Bowen '28). The best results are obtained by the Kolatchev (or Hirschler) method, the Weigl method yielding picutres which are usually distinctly inferior.

2. In some kinds of material, the root-tips of *Equisetum* for example, almost no results are ever obtained except on the osmiophilic platelets. But in other cases, due apparently to a differing cytoplasmic setting, other elements are blackened, sometimes with marked regularity. Of these perhaps the one most commonly impregnated is the plastidome. This term is taken to include the primordial bodies (pro-plastids or archiplasts) in meristem cells from which plastids will later develop, and also fully differentiated plastids of various kinds. The plastidome may be blackened alone (fig. 6), as noted also particularly by Nassonov ('18), but in my preparations of meristem cells the osmiophilic platelets were very frequently demonstrated simultaneously (fig. 3). The Kolatchev method is again the most favorable, but similar results are sometimes obtained by the Weigl method (fig. 5).

3. A very common result, in some kinds of cells it might be said to be a uniform result, the vacuoles (vacuome) may be very clearly demonstrated. This may be effected by an intense blackening of the whole vacuome (figs. 8 and 9), or there may be only an indefinite grayish network marking out the vacuoles (lower part of fig. 4). Sometimes the vacuoles are brought out merely as clear spaces by contrast with a darkened background (upper part of fig. 4). Blackening of the vacuome seems to depend largely upon its contents. In bryophytes, and in pteridophyte root-tips results are very rare, while on certain angiosperm roots constant success is the rule. The method also plays a rôle, and in the case of the vacuome I found the Weigl method most useful. Thus on *Ricinus* root-tips the Weigl method regularly gives results, while the Kolatchev method is relatively useless. In case the vacuome is impregnated, a

simultaneous blackening of the osmiophilic platelets is apt also to occur (fig. 8).

4. Perhaps the most uncommon result is the blackening of those cytoplasmic elements which botanical cytologists frequently call chondriosomes, or 'inactive' chondriome, but which I have temporarily referred to as the pseudochondriome (Bowen '29). These bodies are almost always of a spherical form in meristem cells. Practically the only place where I have found them regularly blackened (by the Kolatchev method) is in the outer plerome of the hyacinth root-tip (fig. 7, which shows also the delicate, thread-like archiplasts). However, they may appear sporadically in other places, for instance the plerome of *Hordeum*, and they are quite regularly blackened in the very old cells of the root-cap of angiosperms at the very apex of the root-tip. In this location, however, the impregnation is not of much use for purposes of study.

5. In addition to these definite cytoplasmic components, cytoplasmic impregnations of a quite different type are commonly found, especially in late prophases and the metaphase of mitosis in meristem cells of angiosperm root-tips. This result was first noticed and fully described by Nassonov ('18). It consists of a very definite blackening of what appears in resting cells to be a cytoplasmic network. In dividing cells the polar caps are also demonstrated as well as the fibers which are attached to the chromosomes (fig. 10). This latter result is most astonishing, and, as Nassonov showed, the number of chromosomes can actually be determined by counting the number of blackened fibers as seen in cross-sections. The whole history of the division figure can be followed, and an exhaustive study of osmic preparations would doubtless yield most interesting results on the nature of the polar caps and other oft-debated features of mitosis in higher plants. When this particular type of blackening occurs, other cytoplasmic elements usually are not demonstrated, but sometimes the plastidome, and in *Hyacinthus* the pseudochondriome, may be in part blackened.

Occasionally, in telophases, the cell-plate may be impregnated, and this may be the only thing blackened in the whole cell (figs. 12, 13). Sometimes the osmiophilic platelets are simultaneously, though inadequately, demonstrated.

6. Impregnation of the nucleus and nuclear structures is a

less common result, but occasionally occurs in a sporadic way in angiosperm root-tips. The whole nucleus may be solidly blackened, with the exception of the nucleolus (lower cell in fig. 11). The nucleolus seems never to be blackened as a whole, but not infrequently one or more granules within the nucleolus are blackened (fig. 11). The exact results vary somewhat but need not here be mentioned in detail, since their meaning is quite unknown. Sometimes a selective blackening of the nuclear membrane is obtained, the remainder of the nucleus being quite unaffected.

7. A very common result especially in cells of the root-cap and central plerome, is the very selective and intense blackening of bodies located within the vacuoles. These are clearly the coagulated masses so often described by Dangeard and Guilliermond. One of the best ways to demonstrate them is by the Hirschler method with brief osmication (4 to 8 days); or better still by fixing according to the Weigl method for 24 hours, and then omitting the osmication step entirely.

8. The osmication methods are known in animal cells frequently to give no result on fatty material. In the case of plant cells, likewise, satisfactory results on the so-called lipoidal granules have not yet been certainly obtained.

9. Here it should be again noted that all sorts of combination results may occur. In these cases, the osmiophilic platelets usually are demonstrated, but not invariably. Thus we may get platelets and plastidome; platelets and pseudochondriome; platelets and vacuome; pseudochondriome and plastidome; platelets, plastidome and pseudochondriome; vacuome and plastidome; vacuome, platelets and plastidome; and still other combinations with the nuclear structures, etc. With some previous knowledge of the cytoplasmic situation in plant cells, however, the analyzing of most of the ordinary impregnation results will present no serious difficulties to the experienced observer.

10. Finally, in the case of counterstaining with acid fuchsin, the resting nucleus and the chromosomes in dividing cells will be regularly stained in red. Preparations in which either the pseudochondriome or the plastidome, or both, have failed to blacken, may, upon proper counterstaining, present these unblackened elements in red.

OTHER NOTES

The results of work with these methods by others are in the main of a similar nature. Nassonov ('18) with the Kolatchev method reported the impregnation of the plastidome alone, and also the blackening of the cytoplasmic network and of the division figure in mitosis. Gatenby ('28) in a recent note corroborates my findings with the Kolatchev method on the osmiophilic platelets. He reports that in addition to the usual Champy fixation in step 1, the following slightly modified fixative also gives very good results:

1 per cent chromic acid 6 per cent potassium bichromate 2 per cent osmic acid	equal parts

The only other worker to report on the osmic methods as applied to plant tissues is Guilliermond ('26, '27). His results include many kinds of cellular components, and so are of particular interest for purposes of comparison with the bulk of this paper, which represents my personal experience. In one important respect his technique differs from that of Gatenby, Nassonov, and myself, and his findings are also different in certain important respects. He used particularly the method of Kolatchev, with one very important change in procedure. He osmicates in 2% osmic acid for 8 to 15 days (or more) at 40°C. Such treatment would ruin any animal tissue I have ever experimented with. As it is, Guilliermond finds that the sections are completely blackened and he then endeavors to differentiate for the cytoplasmic features, particularly the vacuome, by bleaching according to the permanganate-oxalic technique. This method of handling osmic acid is not recommended by any of my own experience, and it may be the reason for some of Guilliermond's anomalous results. Thus he does not mention the osmiophilic platelets, and finds, further, that the pseudochondriosomes are easily and regularly blackened together with the plastidome. His figures of such simultaneous impregnations are certainly not like the appearance of my own preparations. He states that the method rarely blackens the vacuome in higher plants, and then only after an osmication of 15 days. He finds that aleurone grains are beautifully impregnated, and also the vacuome in some of the thallophytes. It is

evident, therefore, that so far as the general possibilities of osmic impregnation of plant tissue are concerned, Guilliermond's results substantiate what I have said about their usefulness. In certain details his results are markedly different from my own and those of Gatenby, but on the other hand his very unusual handling of the osmication step may have been the cause of these differences.

SUGGESTIONS FOR PRACTICE

Osmic impregnation is so erratic in many of its characteristic effects, that one unacquainted with its use would certainly do well to begin with some material where the conditions are known and where a successful result of some kind is reasonably to be anticipated. The best material I know of is the lateral root-tips of *Vicia Faba*, from seeds germinated both in earth and in a moist chamber (Nassonov ('18), Bowen ('28, '29)). Use only the tips of roots not over an inch in length. If the technique is properly carried out, the osmiophilic platelets are almost certain to be demonstrated, together with a variety of other results probably depending on unknown conditions of many kinds. It is my own belief at present that the osmiophilic platelets are the homologue in plant cells of the animal Golgi apparatus. Since it is these bodies which are most regularly impregnated—in some cases they are the only elements to be blackened—the osmic methods are particularly to be recommended for attempting their specific demonstration and further analysis.

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Explanation of plate 1

All of the figures have been outlined as far as possible with the camera lucida at an initial enlargement of approximately 1675 diameters, and subsequently completed free-hand. In reproducing, the figures have been reduced uniformly to an enlargement of approximately 1250 diameters. With the exception of fig. 1, all of the figures are printed with that side of the cell at the top which was originally directed toward the seed (or bulb). The method employed in the preparation of the original object is appended to the explanation of each figure.

Fig. 1. Osmiophilic platelets (blackened rings or rods) in a group of early androgonia from an antheridium of *Polytrichum commune*. Kolatchev method, variation II*b*, 6 days.

Fig. 2. Osmiophilic platelets, from root-tip meristem of *Vicia Faba*. Kolatchev method, variation I*a*, 8 days.

Fig. 3. Osmiophilic platelets (delicate, blackened rings) and plastidome (heavily blackened, elongate bodies), from root-tip (plerome-periblem border) of *Vicia Faba*. Kolachev method, variation II*b*, 7 days.

Fig. 4. Osmiophilic platelets (delicate rings or rods), plastidome (heavily blackened, spheroidal bodies), and vacuome (faintly blackened, above and below the nucleus), from root-tip (plerome) of *Vicia Faba*. Kolatchev method, variation II*b*, 6 days.

Fig. 5. Osmiophilic platelets and plastidome (heavily blackened bodies), from root-tip (root-cap) of *Pisum sativum*. Weigl method, 4 hour fixation, 9 days at 25°C.

Fig. 6. Plastidome, from old root-tip cell of *Pisum sativum*. Kolatchev method, variation I*a*, 7 days.

Fig. 7. Plastidome (delicate threads) and pseudochondriome (intensely blackened granules), from root-tip (plerome) of *Hyacinthus orientalis*. Kolatchev method, variation II*b*, 7 days.

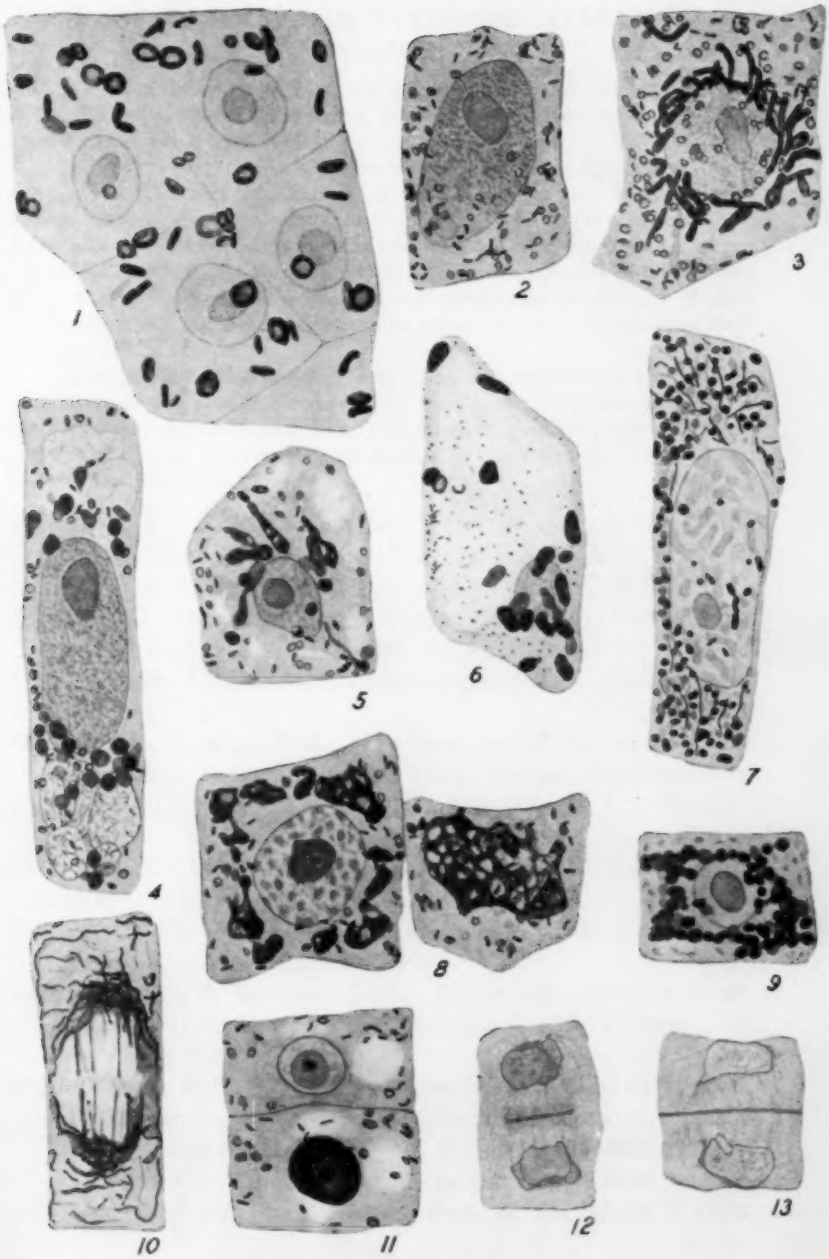
Fig. 8. Osmiophilic platelets and vacuome (blackened net-works), from root-tip (periblem) of *Vicia Faba*. Weigl method, 2 hour fixation, 6 days at room temperature.

Fig. 9. Vacuome (heavily blackened), from root-tip (periblem) of *Ricinus communis*. Weigl method, 1 hour fixation, 6 days at 25°C.

Fig. 10. Cytoplasmic fibrillae, metaphase, from root-tip (plerome) of *Pisum sativum*. Kolatchev method, variation I*a*, 5 days.

Fig. 11. Osmiophilic platelets (delicate rings or rods) and nuclear impregnations, from root-tip (periblem) of *Ricinus communis*. Kolatchev method, variation I*b*, 6 days.

Figs. 12, 13. Cell-plate (telophases), from root-tip (plerome) of *Hordeum vulgare*. Weigl method, 4 hour fixation, 4 days at 25°C.



BOWEN: METHODS OF OSMIC-IMPREGNATION

INDEX TO AMERICAN BOTANICAL LITERATURE
1926-1928

The aim of this Index is to include all current botanical literature written by Americans, published in America, or based upon American material; the word America being used in the broadest sense.

Review, and papers that relate exclusively to forestry, agriculture, horticulture, manufactured products of vegetable origin, or laboratory methods are not included, and no attempt is made to index the literature of bacteriology. An occasional exception is made in favor of some paper appearing in an American periodical which is devoted wholly to botany. Reprints are not mentioned unless they differ from the original in some important particular. If users of the Index will call the attention of the editor to errors or omissions, their kindness will be appreciated.

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